

# ***FBIS Report —***

## **Science & Technology**

***Central Eurasia***

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# Science & Technology

## Central Eurasia

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**Massive Parallelism and Decomposition of  
Algorithms**

*957A0956 Moscow ZHURNAL VYCHISLITELNOY  
MATEMATIKI I MATEMATICHESKOY FIZIKI  
in Russian Jun 95 No 6, pp 988-996*

[Article by V. V. Voyevodin, Moscow; UDC 519.7  
(manuscript received 15 Feb 95)]

[FBIS Abstract] Parallel computers require explicit indication of macroparallelism in algorithm descriptions. This type of parallelism means a large number of arithmetic operations in parallel branches with a small number of links between branches. Traditional forms of writing algorithms, i. e. sequential programs, mathemati-

cal formulas, etc., do not as a rule contain information about macroparallelism. This paper describes mathematical methods to find the macroparallelism in algorithms written in traditional form. Graphs of relationships between information are used to determine links between data. Two key points of the new theory of the information structure of algorithms and programs are presented: formulaic representation of graphs of relationships and formulaic representation of involutes. The information obtained from this analysis can be used to develop autonomous systems of research and program conversion, compilers for parallel computers, portable software, and to design systolic files. References 6: 5 Russian, 1 Western.

**Diffraction of Flat Electromagnetic Wave on Non-Uniform Dielectric Cylinder**

957A0939A *Moscow RADIOTEKHNIKA I ELEKTRONIKA* in Russian May 95 No 5, pp 695-702

[Article by A. S. Ilinskiy, L. M. Nekrasov; UDC 621.396]

[FBIS Abstract] The results of a numerical resolution for diffracting a flat wave on a non-uniform dielectric cylinder are presented and discussed. The conditions applicable for a new numerical algorithm based on a partial Galerkin method are researched. The results which were obtained showed that the method is not receptive to local non-uniformities. However, it is valid for other forms of cylinders besides round ones, such as elliptical cylinders with arbitrary eccentricity, square cylinders, and plates with arbitrary angles of incidence of the flat wave. Moreover, the algorithm was very resistant to counting errors, and calculations could be carried out with acceptable accuracy for even very short waves. 8 figures and 12 references.

**Algorithm of Contrast Signal Detection on Non-Stationary Interference Background**

957A0939B *Moscow RADIOTEKHNIKA I ELEKTRONIKA* in Russian May 95 No 5, pp 784-789

[Article by V. S. Gutin, V. I. Sirotinin; UDC 621.391.26]

[FBIS Abstract] For a locational system that has a receiving antenna with a multi-lobe directional pattern, the task of binary signal detection in a fixed element of resolution is examined, based on delay and Doppler frequency displacement on a background of nonstationary Gaussian interference with an irregular spectrum. The recommended contrast detection algorithm with uniform interference sampling (stationary interference with a regular spectrum) is uniformly the most powerful invariant rule. With nonuniform interference sampling, the algorithm loses the properties of optimality. However, as the results of the analysis showed, its characteristics are sufficiently stable and remain close to optimal at even very steep linear change of the interference amplitude in terms of delay and frequency.

**Impact of Metallic Impurities on Surface Properties of Synthetic Diamonds**

957A0938A *Moscow POVERKHnost: FIZIKA, KHIMIYA, MEKHANIKA* in Russian May 95 No 5, pp 7-13

[Article by G. P. Bogatyreva, K. Yu. Vladimirova, Ye. R. Zusmanov, V. M. Mayevskiy, A. B. Roytsin; UDC 538.971; 539.124143]

[FBIS Abstract] Samples of diamonds were subjected to thermochemical processing and hydrogenation at temperatures of 1200 K to 1500 K. Electronic paramagnetic resonance spectra were studied, as well as magnetic susceptibility, dielectric losses, wetting ability, and composition of the impurities. It was shown that ferromagnetic inclusions markedly affect the bulk and surface properties of synthetic diamonds. Their presence can particularly promote hydrogenation of samples and reduce the hydrophilicity of their surfaces.

**Effect of Thermal Annealing on Electrical Characteristics of Metal-Dielectric-Gallium Arsenide Structures**

957A0938B *Moscow POVERKHnost: FIZIKA, KHIMIYA, MEKHANIKA* in Russian May 95 No 5, pp 18-26

[Article by V. I. Gaman, V. M. Kalygina, A. V. Panin, T. P. Smirnova; UDC 537.311.33:621.382]

[FBIS Abstract] The effect of thermal annealing on *C-U* and *G-U* properties of the Pd-dielectric-*n*-GaAs structure is investigated. BN and SixNyOz films grown by the plasmachemical method were used as the dielectric. It was shown that changing the electrical characteristics of these structures depends upon the conditions of synthesis and the mode of heat treating. It is supposed that the studied effect may be explained by the interaction of hydrogen with defects in the surface layers of a semiconductor near the dielectric-GaAs interface.

**Optic Absorption in Ion-Implanted Gallium Arsenide**

957A0938C *Moscow POVERKHnost: FIZIKA, KHIMIYA, MEKHANIKA* in Russian May 95 No 5, pp 27-30

[Article by Yu. A. Danilov; UDC 537.311.33]

[FBIS Abstract] In the 0.8-1.2  $\mu\text{m}$  wavelength range, the optical transparency of gallium arsenide irradiated with hydrogen and silicon ions with energies from 40 - 100 keV was investigated. It was shown that the magnitude of the  $a_b d_b$  absorption exponent rises as the amount of ions increases, and reaches saturation during

amorphization at a photon energy level of  $> 1.2$  eV. The absorption coefficient for gallium arsenide amorphized by ion implantation changes insignificantly when the energy is varied or the type of ions is changed, and averages  $\alpha b \ln F 5 \times 10^4 \text{ cm}^{-1}$  at a wavelength of  $1.13 \mu\text{m}$ . At a photon energy of  $> 1.2$  eV, the absorption exponent will not become saturated right up until a dosage of  $10^{16} \text{ cm}^{-2}$ . It is supposed that amorphization of the gallium arsenide is associated with the accumulation of *EL2*-type defects. An assessment of *EL2* saturation concentration gives a value of  $-2 \times 10^{21} \text{ cm}^{-3}$ .

**Effect of Ion Irradiation on Structural Characteristics of  $\text{ZrO}_2$  Films on Silicon Substrate and Electrophysical Properties of  $\text{GaAs}/\text{SiO}_2$  Surface**

957A0938D Moscow POVERKHINOST: FIZIKA, KHIMIYA, MEKHANIKA in Russian May 95 No 5, pp 96-98

[Article by V. A. Kamin, D. O. Filatov, I. A. Karpovich, R. I. Bochkova; UDC 539.22]

[FBIS Abstract] Crystalline forms of dielectric films of zirconium oxide stabilized by yttrium (YSZ) were irradiated by plasma energy particles and the results were investigated, along with the properties of the surface of the  $\text{GaAs}/\text{SiO}_2$  interface. The analyzed data showed that in ion plasma sputtering devices, the deposited film and semiconductor substrate are subjected to ion irradiation, whose impact can be seen in the change in the structure of the films and the density of the state on the surface of the semiconductor.

**Research on Laser Pulse Effects on  $\alpha\text{-Ge}/\text{Si}$  Structure by Rutherford Backscattering and Ion Channelling Method**

957A0938E Moscow POVERKHINOST: FIZIKA, KHIMIYA, MEKHANIKA in Russian May 95 No 5, pp 140-144

[Article by E. V. Monakhov, K. K. Burdel, N. G. Chechenin; UDC 537.534;548.4]

[FBIS Abstract] The Rutherford backscattering and ion channelling method was used to investigate structural and compositional changes in the  $\alpha\text{-Ge}/\text{Si}$  system with laser pulse effects. The germanium diffusion coefficient is assessed in a melt, and the conditions for the epitaxial growth of  $\text{Ge}_x\text{Si}_{1-x}$  films on a silicon substrate with  $111$  orientation are determined. As a result of mixing the  $\text{Ge}/\text{Si}$  layers, followed by recrystallization of the melt, it is demonstrated that it is possible to create a sufficiently perfect epitaxial layer of  $\text{Ge}_x\text{Si}_{1-x}$  even in relatively unfavorable epitaxial conditions on the surface. The most perfect layers are obtained by using multiple pulse irradiation. If single irradiation is used, a  $\text{Ge-Si}$  layer developed with a strongly alternating composition. Further, the substituting fraction of germanium sharply dropped when the concentrations of germanium were higher than critical, which are approximately 4% when  $W = 2.3$ .

**German Credits for Development of Russian Communication Network**

957A0999 Moscow ELEKTROSVYAZ in Russian  
Mar 95 No 3, p 39

[Article by K. D. Bushe, G. Khaas, Alcatel SEL RFT]

[FBIS Translated Text] Alcatel is one of the world's largest producers of telecommunications equipment. Alcatel has been successful in the markets of many countries. Its subsidiary in Stuttgart (Germany), Alcatel SEL, has its on subsidiary, Alcatel SEL RFT, that traditionally provided communication equipment to the former USSR.

Alcatel SEL RFT was created on the foundation of Nachrichtenelektronik, a national industrial complex, which up until 1989 was the main exporter of telecommunications equipment to the countries in the socialist camp. During 1962-1990 it supplied analog and semi-electronic dial offices with a total capacity of seven million connecting and subscriber lines, 4100 transmission systems with pulse code modulation, 100 million kilometers of coaxial cable, and almost 0.5 million electro-mechanical and electronic teletypes. Moreover, there was a demand for shortwave transmitters for ships and radio communication systems for the railroad.

Its excellent knowledge of the Russian market and local communication networks, as well as features of its commercial activity, permitted Alcatel SEL RFT, together with Alcatel, to become a reliable partner of their colleagues from the CIS nations as providers of telecommunications equipment. Alcatel SEL RFT considers Russia an extremely promising market and is undertaking its own work here based on agreements with the Ministry of Communication of the Russian Federation and in cooperation with regional Elektrosvyaz joint stock companies.

Each year, Alcatel SEL RFT implements projects in Russia with a value of over 100 million DM. These projects are funded by credits from the German government, based on agreements with the Federal Oversight Department of Germany. They are also funded by local Russian sources. An important pre-condition for successful completion of these projects is certification of all the equipment introduced by the firm by the appropriate organizations of the Ministry of Communication of the Russian Federation in Moscow and St. Petersburg. Moreover, the Technical Center of Alcatel SEL RFT and its St. Petersburg joint venture, Alcatel DS, are creating an effective service system which has regional divisions at various sites.

Alcatel SEL RFT is striving to work with the program for the development of the telecommunications network of Russia by developing concepts of collaboration and financing, as well as through its independent participation in projects. This policy corresponds to market demands. The situation is such that, on the one hand, Russian industry cannot yet quantitatively or qualitatively satisfy the country's need for modern communication equipment, and on the other hand, the foreign partner should supply nothing more than simple high-performance equipment.

An important condition of implementation of the project is agreed concepts of financing, since only rarely is direct payment in hard currency made. Government credits, credits of the World Bank and the European Bank of Reconstruction and Development, liquidated with the help of foreign communication operators, as well as barter deals, represent only some of the practical forms of financing of the provision of equipment by Alcatel SEL RFT to Russia. Along with partners from Perm, Nizhny Novgorod, Volgograd, Chelyabinsk, Stavropol, Orenburg, Tula, Tambov, Arkhangelsk, Bryansk, Yoshkar-Ola, Saransk, Penza, Syktyvkar, Ulyanovsk, Smolensk, Ivanovo, Vladimir, Vladikavkaz, Kaliningrad, and Sverdlovsk oblast, the firm is implementing a program to introduce an Alcatel 1000 S12 digital switching system with a total capacity of 297,000 numbers and 160,000 connecting lines.

In each specific case, a separate financing plan is being developed, because the leading role is played by export operations based on government credit under the guarantee of the German insurance company Hermes, as well as agreements signed with the Federal Oversight Department of Germany. Among the latter are the following:

- a basic credit-goods agreement reached by the Oversight Department of the Federal Republic of Germany and the communication administration of the corresponding oblast;
- a contract to provide equipment in which Alcatel SEL RFT is the exporter on the German side, and the Russian importer is some communication administration or operator;
- a long-term contract on cross deliveries: the Russian side is already acting as an exporter, and some Western European buyer of Russian goods will be the importer;
- a contract on the financing of cross deliveries: in this agreement the corresponding communication administration or operator pays some Russian enterprise in rubles the cost of the goods they

deliver to the German market according to the previous contract;

- an interbank agreement reached by one of the German banks, acting as creditor, and one of the Russian regional banks acting as a borrower.

This scheme, which provides for simultaneous execution of the five contracts mentioned above, guarantees payment for communication equipment by direct cross delivery of Russian goods, and provides Russian communication administrations and operators with the opportunity to expand and modernize networks on a non-hard currency basis.

However, a necessary condition of the implementation of this type of scheme is the availability of goods for cross delivery with a demand in Western Europe. This condition also defines the limits of this type of financing. Nonetheless, the associated secondary expenditures are compensated by fast construction of a modern digital telecommunication system, without which it would be impossible for a market economy to exist and which in turn, guarantees the operators profits in the shortest possible time.

Alcatel SEL RFT currently has orders from clients in the CIS nations totaling about 1 billion DM. Along

with the development of the concepts of financing, the company, to increase its competitiveness, is striving to increase its contribution to the creation of a production base for Alcatel equipment in Russia, including the design of equipment, its installation and start-up, and the production of system components.

The organization of a network to service equipment installed in the aforementioned oblasts of Russia involves competent local technical centers, ever increasingly staffed by Russian specialists trained in Germany who have studied the hardware and software of the Alcatel 100 S23 digital switching system. That is one of the forms of the transfer of know-how that is increasing the interest in long-term collaboration with Alcatel.

The practice of close collaboration between specialists of Alcatel SEL RFT, who are expertly informed on the Russian market, and their clients distinguishes the firm from other companies. Alcatel SEL RFT is continuing the search for optimal financing concepts, and will strive to be a reliable partner for customers from Russia in the modernization and development of local communication networks. The introduction of Alcatel equipment is an excellent recommendation for the firm, and satisfied clients are the best advertisement.

**Would It Be More Expensive to Erect Fast Reactors Than Thermal Ones Under the Present State of Russian Nuclear Power Engineering?**

957A1027A *Moscow ATOMNAYA ENERGIYA*  
in Russian Jan 95 No 1, pp 3-8

[Article by M. F. Troyanov, V. M. Poplavskiy, V. S. Kagramanyan, A. V. Malenkov (FEI), A. I. Kiryushin (OKBM), K. L. Suknev and V. N. Yershov (SPBI AEP) (acronyms not further identified); UDC 621.039.526.003]

[FBIS Translated Text] From the very beginning, the main goal of creating fast reactors was to breed plutonium and ensure an optimistic rate of development of nuclear power engineering in the face of limited natural uranium resources. Now, because of critical reconsideration of the forecasts for development of nuclear power engineering in the direction of slowing its pace, the objective of breeding plutonium has lost its urgency, and has been postponed to a more distant future. At the same time new problems have made themselves known: In the short term—dealing adequately with energy-producing plutonium, 30 tonnes of which have already been accumulated by the Mayak Production Association as a result of chemical processing of uranium fuel at the RT-1 plant, and in the long term—dealing with long-lived radiotoxic actinoids accumulated and accumulating in the spent fuel of thermal reactors. In addition up to 100 tonnes of weapon-grade plutonium are expected to be released for use as a result of disarmament, and its optimum use is another task facing nuclear power engineering.

Estimates made both here and abroad show that fast reactors can be reoriented on solving new global problems—reusing the accumulated plutonium (without breeding) and burning off radiotoxic Np, Am and Cm. In particular, energy-producing plutonium accumulated at RT-1 and released weapon-grade plutonium could be reused by a complex consisting of a plant producing uranium-plutonium fuel and three or four of the BN-800 reactors of the Uralskaya and Beloyarskaya AES. These aspects of new stimuli for developing fast reactors have been studied and discussed in detail in works such as [1,2].

Construction of a small series of fast reactors would make it possible on one hand to ensure an economically acceptable scale of fuel production, and on the other, to solve the problem of free plutonium within in a short period of 20-30 years by transforming it into spent fuel.

At the same time further development of the fast reactor program can now be justified by the possibility such reactors have for competing commercially in the nuclear power engineering system with newly developed light-water reactors, on the condition that safe operation can be achieved. As always, capital outlays are the primary concern.

Two peculiarities can be noted about development of Russian nuclear power engineering in the present stage. First of all, there is the particularly Russian circumstance that the safety of series VVER and RBMK thermal reactors in their present form must be raised to satisfy recommendations accepted worldwide. Second, plans for a nuclear power plant equipped with the BN-800 have already passed all the necessary expert examination, and on the whole they satisfy current requirements of technical standards. Moreover ways have been found to improve the technical and economic characteristics of fast reactors while maintaining the high safety level that has been attained, or even surpassing it. These features need to be considered when considering the competitiveness of nuclear power stations with fast reactors in the overall structure of nuclear power engineering.

Forecasts made abroad reveal a tendency for the economic characteristics of fast power reactors to approach those of thermal reactors upon transition to series construction. The first experimental industrial fast reactors were inferior to light-water thermal reactors both in unit capital investments and in the cost of the electric power produced. In our country the BN-600 was 1.5 times inferior to the main VVER-1000 block at the Novovoronezhskaya AES [3]. A figure of 2.5 was cited in the West in a comparison of the Superphoenix reactor with a series-manufactured PWR thermal reactor. This is explained by differences in the degree of industrial assimilation of the compared reactors and the fuel cycle.

Plans for the EFR fast reactor (European fast reactor) with an electric power capacity of 1,500 MW have been drawn up through the joint efforts of Western European countries. Owing to improvements, structural metal content was significantly reduced. As a result the unit capital investments should decrease 45 percent in comparison with the Superphoenix-1, and then another 22 percent upon transition to series manufacture. Table 1 gives forecasts of possible economic characteristics of series-manufactured EFR and French PWR reactors of the future (series No 4).

Table 1. Economic Indicators of Reactors of the Future [4]

Characteristic	EFR	PWR
Components of the cost of electric power, francs/kWbhr:		
capital	13.5	10.7
operating	4.4	3.9
fuel	4	5.1
Cost of electric power, francs/kWbhr	21.9	19.7
Ratio of indicators, EFR/PWR:		
capital components	1.26	
cost of electric power	1.11	

Forecasts show that unit capital investments into series-manufactured fast EFR reactors will be US \$1,800-2,000, while those of thermal light-water reactors will be \$1,500, which corresponds to the ratio of 1.26 in Table 1 for the capital components of the cost of electric power.

In the general case, the capital outlay ratio will always depend on the country for which this comparison is made, and on the particular site and the number of blocks it contains. The 1.26 figure pertains to a comparison with a French PWR. If we were to make a similar comparison for the conditions of Germany and Great Britain, the corresponding figures would be 1.16 and 1.1.

In our country, operation of experimental industrial BN-350 units since 1972, and especially of BN-600 units since 1980, allowed us to accumulate design and operating experience. The 14-year experience of operating

the BN-600 fast reactor, which has a promising integral (tank-type) layout, made it possible to accumulate comprehensive experience in operating reactor plant equipment, and to account for this experience in the design of the BN-800 (Table 2). The economic characteristics of the plan of the BN-800 were better chiefly because when the BN-600 was developed as an experimental industrial model, a margin was designed into all parameters. The capacity of the BN-800 was increased with almost no change in the absolute materials consumption of the reactor plant. Mention should also be made of the tendency for the relative materials consumption of the BN-800 plant to approach the same indicator of the series-manufactured domestic thermal VVER-1000, which is 8.44 tonnes/MW (electric). A new development of the OKBM—the BN-600M—has significantly lower materials consumption due to a decrease in the number of heat exchange loops and use of an integral (shell-type) layout for the steam generator [6].

Table 2. Comparative Characteristics of BN and VVER Reactor Plants (Data of the OKBM)

Indicator	BN-600	BN-800	BN-600M	VPBER-600*
Thermal capacity, MW	1470	2100	1520	1800
Electric power at generator terminals, MW	613	793	647	640
Efficiency (gross), %	41.7	37.7**	42.5	35.5
First loop (kontur) heat-transfer agent	Sodium	Sodium	Sodium	Water
Number of heat exchange loops	3	3	3	2
Number of heat exchange loops (petli)	3	3	2	4
Core fuel assembly life, effective days	480	420	600-750	1000-2700

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Indicator	BN-600	BN-800	BN-600M	VPBER-600*
Average fuel depletion, MWlbday/kg	60	66	80-100	51
Reactor weight, tonnes	7875	8435	6725	6085
<b>including:</b>				
reactor with internal equipment***	3500	3060	2490	3850
first loop and equipment cooling loop	2820	3620	2368 (968)	376
external fuel assembly overload system and special devices for equipment replacement	634	634	549	416
emergency reactor cool-down system	120	205	360	1123
heat-transfer agent cleaning system	126	107	90	320
metallic structures unique to a fast reactor	675	810	870	-
Specific materials consumption of reactor plant, tonnes/MW	13	10.5	10.4 (8.23)	9.7

\*Characteristics of the VPBER-600 are for the conceptual design of an integral reactor layout.

\*\*Efficiency (gross) is given for a thermal system with two low-pressure cylinders.

\*\*\*The weight of external mechanisms of the fuel assembly overload system and special devices accounted for in the row "External fuel assembly overload system" is not included with the reactors.

Data for the version with a shell-type steam generator are given in parentheses.

Safer thermal reactors of a new generation are currently being planned on the basis of domestic construction experience. Furthest along are plans for the moderate-capacity NP-500 (loop [petlevoy]) and VPBER-600 (integrated) reactors. In this case the increase in safety requirements brings the economic indicators of fast and thermal reactors closer together, inasmuch as these goals had been set in fast reactors previously, and were embodied in the plans, and some of them are attained with simpler resources owing to low pressure, effective heat output, a high level of natural circulation etc. The specific materials consumption of the VPBER-600 reactor is noticeably greater than that of the series-manufactured thermal reactor, and comes closer to that of a reactor plant containing fast reactors (BN-800).

It should be noted that further development of fast reactors will involve technical developments such as reducing the number of heat transfer loops by introducing passive safety equipment and an integrated (shell-type) steam generator layout, and making the automated pro-

duction process control system less expensive. Of course, the final choice will be based on the overall selection criteria.

If we consider series manufacture as a factor significantly determining the technical and economic parameters of nuclear power stations, then from a historical standpoint fast reactors have always been at a disadvantage. The reason for this is that every new modification of such a nuclear power station has been unique, at the same time that other types of reactors, for example the VVER, were already being built in series. Initial planning of the Yuzhno-Uralskaya AES, where a small series of blocks equipped with the BN-800 is to be built, provided an opportunity for evaluating the effect of the series factor in a comparison of different types of nuclear power stations. It is evident from Table 3 for example that erection of three BN-800 blocks at the Yuzhno-Uralskaya AES and three NP-500 blocks at the Kolskaya AES is almost equivalent in unit capital investments.

Table 3. Cost of Building Nuclear Power Stations With Thermal Reactors of a New Generation and Fast Reactors (in Prices of the Fourth Quarter of 1992)

AES, Generation, Block	Electric Capacity, MW	Capital Investments, million rubles	Unit Capital Investments, rubles/kW (el.)	Unit Capital Investments Relative to Series-Manufactured NP-500
<b>Yuzhno-Ural'skaya AES:</b>				
1st generation, BN-800, pilot	800	56,694	70,860	1.45
2d generation, 2 BN-800, series	1,600	55,770		
Total	2,400	112,464	46,860	0.96
Beloyarskaya AES, 3d generation, BN-800, series	800	44,949	56,186	1.15
Kolskaya AES, 3d generation, 3 NP-500, series	1,890	92,400	48,888	1
Sosnovyy Bor, NP-500, pilot	630	36,795	58,404	1.19
Primorskaya AES (Dimitrovgrad), 1st generation, VPBER-600, pilot	1,200	70,950	59,125	1.21

It would be interesting to examine the economic aspects of different energy sources for the same service area. The degree to which the particular region has been developed and its readiness to erect a facility are excluded from the discussion in this case. Thus, the St. Petersburg Atomenergoprojekt Institute drew up different versions of an energy source for Kaluga Oblast (Table 4). Because the calculations were made several years ago, we are concerned with the relative and not

the absolute financial figures. It is evident from Table 4 that the NP-500 thermal reactor and the BN-600M fast reactor are almost equally economical in terms of the cost of produced electric power, and are significantly more economical than a coal-burning thermal electric power station. It may be concluded from Table 5 that thermal and fast reactors are equally economical, and are significantly more effective than a coal-burning thermal electric power station.

Table 4. Technical and Economic Indicators for Different Versions of an Energy Source for Kaluga Oblast, in 1991 Prices and Relative to a Nuclear Power Station Equipped With the NP-500

Electric Power Station	Design Electrical Output, MW	Capital Investments Into Construction		Operating Expenses, million rubles/year	Cost of Electric Power	
		million rubles	relative units		kopeks/kWbhr	relative units
3NP-500	1935	2317	1	362	2.87	1
3BN-600M	1947	2069	0.89	342	2.72	0.95
Coal-burning thermal electric power station	2100	1881	0.81	799	6.33	2.21

**Table 5. Comparative Cost of Construction at the Site of the Beloyarskaya AES (data from the St. Petersburg Atomen-ergoprojekt Institute), thousand rubles (1991 prices)**

Indicator	AES With One BN-800	AES With Two NP-500*	Beloyarskaya TES With Two Coal-Burning 500 MW Blocks
Construction and installation	668,928	553,985	736,100
Equipment	583,638	1,573,995	555,200
Other outlays	224,505	244,050	160,600
Total	1,477,071	2,376,030	1,451,900** 6,371,900***
Relative cost, rubles/kW	1,846	1,842	1,451** 6,372***
Relative cost compared to NP-500	1.02	1	0.77** 3.46****

\*Installed electric capacity of a 645 MW NP-500.

\*\*, \*\*\*\*Respectively without and with regard for the cost of a rail approach line.

\*\*\*Cost of the rail approach line.

Consequently the economics of any type of nuclear power station depend to a greater degree on the place of construction, the degree to which the site is developed, and the number of power generating blocks on it. These factors assume the forefront in a comparison of fast and thermal reactors, eclipsing the economic characteristics typifying the reactors *per se*. Considering the real economic situation in Russia, we doubtlessly need to exercise caution in considering the absolute values of the economic parameters of nuclear power stations analyzed here. Nonetheless, their relative comparison allows us to discern the trends in development of the main types of reactors for nuclear power engineering.

Thus present circumstances are such that development of fast reactors now pursues new strategic goals in addition to the traditional one of economizing on natural uranium: reusing produced energy-producing plutonium and released weapon-grade plutonium in the short range, and burning off Np, Am and Cm in the long range. Analysis of the development of nuclear power engineering in different countries shows that the technical and economic indicators of nuclear power stations equipped with light-water and fast reactors with a sodium heat-transfer agent are tending to even out. This is being brought about to a significant extent by the stiffer safety requirements, in which case, given what it would take to satisfy them, this could be done with simpler equipment in the case of a fast reactor.

Transition to series construction is an important factor of improving the economic parameters of nuclear power stations with fast reactors. For example, in this case the

economic indicators of a planned nuclear power station equipped with the BN-800 come close to those of a nuclear power station with the VVER-1000, and they are not inferior to those of a station equipped with the NP-500. At the same time using both types of reactors located at the same site is significantly more effective than using a coal-burning thermal electric power station.

Certain engineering solutions satisfying present and future safety requirements make further improvement of the technical and economic characteristics of fast reactors possible.

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**Data Analysis and Physicochemical Modeling of the Radiation Accident in the Southern Urals in 1957**

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[Article by S. A. Kabakchi, A. V. Putilov (State Scientific Center of the Russian Federation Physicochemical Scientific Research Institute imeni L. Ya. Karpov) and Ye. R. Nazin (Military Academy of Chemical Protection imeni Marshal S. F. Timoshenko); UDC 621.039.7]

[FBIS Translated Text] Major radiation accidents at enterprises involved in the nuclear fuel cycle are a unique source of information that can have a decisive influence on the development of nuclear- and radiation-safe plants and establishment of a system of safety standards. As a rule because there is so little objective information about radiation incidents and accidents, it is difficult to determine the true physical cause of a physicochemical phenomenon that developed at some stage of the production process at an uncontrollable rate or in an undesirable direction.

This article shows the possibility for revealing the causes of a radiation accident when information is insufficient by analyzing indirect data on its consequences. The radiation incident at the waste dump of a radiochemical plant in 1957 was the object of analysis. The article does not address the ecological consequences of the accident. Emphasis is laid on revealing the essence and laws governing development of the physicochemical processes that led to the accident and were the cause of dispersal of radionuclides over a large distance. As far as we know, this problem has not been discussed in the literature.

**Analysis of Data on the Accident**

The accident in the Southern Urals in 1957 attracted the attention of the world community after publication of an article in which it was concluded, on the basis of an analysis of open publications reporting research on the effects of radiation upon animals and plants of the Uralsk vicinity, that a massive release of radionuclides into the environment (a minimum of 50 megacuries and a radioactive contamination area of 400 to 900 km<sup>2</sup>)

occurred in late 1957 or early 1958 in the vicinity of Kyshtym, Chelyabinsk Oblast. This conclusion created a sensation because Western experts (including intelligence services) knew nothing about a radiation accident of such a scale. Various suppositions regarding the causes of the accident were offered in a series of publications [2-12] following this report: radioactive fallout from thermonuclear weapon tests on Novaya Zemlya; deliberate dropping of an atomic bomb with a yield of around 20 kilotonnes (TNT) to determine the stability of the radiochemical plant and its infrastructure; a sudden release of radionuclides resulting from a self-sustaining nuclear chain reaction in storage trenches containing radioactive wastes from weapon-grade plutonium production; an accident occurring during assembly of a nuclear weapon; a steam or chemical explosion in a radioactive waste dump; the result of many successive releases of small quantities of radionuclides.

It was noted in [9] that hypotheses regarding exotic processes such as accidental (or intentional) explosion of a nuclear device or a reaction arising due to selective adsorption of plutonium by slurry in a waste trench are not necessary to explain the release. Radiochemical production wastes with a high relative radioactivity are chemically active systems that can boil as a result of intensive heat evolution. Because potentially explosive organic nitrates are used in some stages of radiochemical processing of irradiated uranium, scenarios involving chemical and steam explosions, fire, rupture of waste storage units and so on may be written. A chemical explosion in a liquid waste dump would cause formation of a large number of droplets that would then be dispersed and undergo evaporation in response to the heat of radioactive decay. Good conditions are created in this case for formation of aerosols containing radioactive uranium fission products. If a fire accompanies the explosion, the flow of heated air will raise dispersed aerosol particles to a considerable altitude.

The chemical explosion hypothesis can explain the large area of terrain that was observed to be radioactively contaminated, and it does not require any additional assumptions. Accidents of this type did happen at chemical enterprises before.

The possible physicochemical causes of the accident were discussed in [10]. An explosion in a storage dump containing highly radioactive wastes from acquisition of weapon-grade plutonium was recognized to be the most probable cause of the release and of contamination of the terrain. The authors of this paper suggested (and it is now known for sure [13]) that in the 1960s the USSR used an acetate procedure to process spent uranium (precipitation of uranyl and sodium acetate). A concentrate consisting of fission products from the first

cycle of division, which had a relative radioactivity of 10-100 Ci/liter, was stored in underground containers in liquid form for 3-5 years. After its radioactivity dropped below 1 Ci/liter, it could be stored in other ways (solidification, burial in deep subterranean horizons etc.). The following physicochemical processes could have caused release of a large quantity of fission products into the atmosphere: a huge chemical explosion or fire after a micro-scale self-sustaining chain reaction causing superheating of the wastes; ignition of readily flammable solvents they contained; explosion or detonation of old nitrate-containing wastes; detonation of radiolytic hydrogen, and a steam explosion (due to generation of a large quantity of heat during radioactive decay) in a storage container at the radiochemical plant. The authors of [10] explained the abnormally low  $^{137}\text{Cs}/^{80}\text{Sr}$  radioactivity ratio, which is atypical of the highly radioactive wastes produced by the procedure for obtaining weapon-grade plutonium, by the fact that  $^{137}\text{Cs}$  was isolated from the wastes by the ammonium alum procedure (apparently a procedure for isolating cesium using ammonium phosphomolybdate). What makes this procedure significantly different from others is presence of a sizable quantity of ammonium nitrate in the wastes—a substance capable of spontaneous explosion and detonation (catastrophic explosions have been known to occur with fertilizers containing ammonium nitrite).

As a result the following scenario of events leading to the radionuclide release was composed in [10]: Due to damage to the cooling system or to other safety systems of a storage facility containing highly radioactive wastes, slurry or the solid precipitate in a container became superheated, causing an explosive reaction between nitrates and organic ingredients of the wastes (acetates or solids), or detonation of ammonium nitrate. In this case the energy release could have been large: If a 1,000-3,000 m<sup>3</sup> container was two-thirds full of ammonium nitrate with a concentration of 2 to 8 moles/liter, the explosion would have been equivalent to 0.1-1 tonnes of TNT. Because the waste containers are stored in groups of several units, an explosion of one of them could have caused detonation of others, which could in principle have contained wastes of a different type with a different radiochemical composition. This in the opinion of the authors could explain the unusual composition of the release.

The major radiation incident that occurred in the Southern Urals on 29 September 1957 is described in detail in [11-13]. Following isolation of weapon-grade plutonium, liquid wastes were transferred at the radiochemical plant of the Mayak Scientific-Production Association into storage tanks with a volume of around 300 m<sup>3</sup>, made from stainless steel and situated in concrete-

lined trenches dug slightly below the ground surface. The storage tanks were around 2 km from the plant. To prevent situations that could cause a chemical explosion in the wastes, the containers were cooled by means of heat exchangers on the interior wall of the storage facility. The design of the heat exchangers did not allow for their repair in case of damage. In 1956 the heat exchanger of one of the storage tanks was shut down due to a fault. Calculations carried out by plant specialists showed that the wastes would remain stable even in the absence of cooling (as was noted in [12], their calculations were wrong). As a result no attempt was made to set up heat removal from this container for over a year. Water began evaporating from wastes in the tank with the disconnected heat exchanger, and explosive nitrates and acetates concentrated on the interface between the wastes and the air. A chance spark from a faulty piece of monitoring and measuring equipment caused detonation of the salts. The explosion ruptured the container and ejected its contents.

In the opinion of the author of [13] the force of the explosion of the nitrate and acetate mixture in the storage facility was equivalent to 5-10 tonnes of TNT. This caused release of 70-80 tonnes of wastes containing radionuclides with a radioactivity of around 20 MCi. Approximately 90 percent of this quantity fell out near the place of the accident. The remaining 2 MCi formed a cloud 1 km high, which traveled over a significant amount of territory. It is estimated that 15,000-20,000 km<sup>2</sup> were contaminated above 0.1 Ci/km<sup>2</sup> within a radius of 300 km.

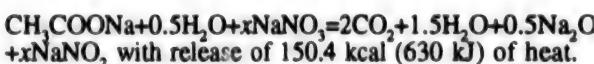
#### Modeling the Accident Process

Hypotheses regarding the causes of the radiation accident in the Southern Urals based on an analysis of indirect data were compared in the 1970s with actual data [11,12]. The comparison showed that in qualitative respects these hypotheses faithfully reflected the essence of the processes occurring during the accident. The results of modeling the explosion according to a scenario presented in [10-12] are presented below. They permitted estimation of the energy of the explosion and the height of release of the radionuclides.

Exothermic processes begin in mixtures of sodium nitrate and solid organic materials at a high temperature (360-400° [14]). This is because organic matter interacts with oxygen—a decomposition product of sodium nitrate, rather than with this compound itself. Pure sodium nitrate breaks down into nitrite and oxygen at 380°C. The sodium nitrite that forms as a result is stable. Mixtures weighing around 100 mg consisting of 30 percent sodium acetate and 70 percent sodium nitrate by weight, and 10 percent sodium acetate and 90 percent sodium nitrate (with molar ratios of 1:3.74 and

1:14.6 respectively) were subjected to derivatographic [?: derivatograficheskoye] analysis. It was shown that after removal of water (around 10 percent weight loss in the first case and 5 percent in the second) at temperatures of 385 and 370°C respectively, an intensive exothermic process (with 620 kJ/kg heat release) coupled with a weight loss of 15 percent begins in the first case, and a relatively slow exothermic process (with 83 kJ/kg heat release) coupled with a weight loss of 2.1 percent begins in the second.

Considering the data of derivatographic analysis, the processes that actually occur when storage mixtures undergo heating, and the fact that boiled-down highly radioactive wastes are a stoichiometric mixture or one with excess oxidizer, the most probable equation for the reaction between the mixture's components would be:



Data from derivatographic analysis indicate that only part of the heat is actually released upon interaction of the mixture's components, depending on the composition of the mixture. In the first approximation the relationship of heat release  $Q$ , to the composition of the mixture (the ratio  $x$  of the number of moles of sodium nitrate to the number of moles of sodium acetate) when  $x \neq 4$  may be expressed as  $Q = 822.6 - 50.6x$  kJ/kg. The quantity of gases formed per kilogram of sodium acetate and nitrate mixture is  $V = 22.4m/M$ , where  $m$  is the number of moles of gaseous reaction products, and  $MM$  is their molecular mass. Using the ratio  $x$  for the sodium acetate and nitrate mixtures, and with regard for data from derivatographic analysis, when  $x \neq 4$  we have  $V = (560 - 22x)/(1 + 1.036x)$  cubic nanodecimeters per kilogram.

The TNT equivalent is  $\text{TNT}_{\text{equiv}} = Q/4520$ ; for these mixtures,  $\text{TNT}_{\text{equiv}} = 1.1 \times 10^{-2}(16.25 - x)$ . The equivalent charge is  $W = 1.1 \times 10^{-2}m_{\text{equiv}}/(16.25 - x)$ , where  $m_{\text{equiv}}$  is the weight of the mixture boiled down to dry state, tonnes.

According to data in [15], change in pressure upon explosion in a leak-tight volume depends on the ratio of the weight of the charge  $W$  to the volume of the chamber  $V$ . When  $W/V \leq 10 \text{ kg/m}^3$ , this relationship is described approximately by the equation  $P_{\text{expl}} = 1300(W/V)^{0.755}$ , where pressure  $P_{\text{expl}}$  is measured in kPa, and  $W/V$  is measured in  $\text{kg/m}^3$ . For these mixtures and when  $x \neq 4$ , in the case of an explosion  $P_{\text{expl}} = 1300[1.1 \times 10^{-2}m_{\text{equiv}}(16.25 - x)/V]^{0.755} \text{ kPa}$ .

In the case of a ground burst, the maximum height to which the products are lifted is  $H_{\text{max}}^{\text{ground}} = W^{0.2} \text{ km}$ , where  $W$  is the weight of the TNT charge in tonnes. In our case of a partially underground explosion  $H_{\text{max}}^{\text{under}} = 0.85H_{\text{max}}^{\text{ground}}$  or, expressing the energy of explosion of a sodium nitrate and acetate mixture by the TNT equivalent,  $H_{\text{max}}^{\text{under}} = 0.85[1.1 \times 10^{-2}m_{\text{equiv}}(16.25 - x)]^{0.2} \text{ km}$ .

The results of calculating the parameters of explosion of several different mixtures, presented in the table, show that at the molar ratio of sodium acetate and nitrate characteristic of highly radioactive wastes and at a boiled-down salt weight of 30 tonnes, the equivalent charge is 2.4-4.1 tonnes of TNT. The power of such a charge is sufficient to generate a pressure of up to 10 MPa in the container, and to eject its contents to a height of 1 km. The power of the equivalent charge coincides in order of magnitude with estimates in [13].

Parameters of Explosion of a Mixture of Sodium Nitrate and Acetate (Weight of Boiled-Down Salts—30 tonnes)

Composition of Mixture, % by weight		Heat Release, kJ/kg	Quantity of Gases Per 1 kg of Mixture, nm <sup>3</sup>	TNT Equivalent	Explosion Pressure, kg/cm <sup>2</sup>	Height of Cloud, km
NaAc	NaNO <sub>3</sub>					
19	81	620	92	0.137	94	1.12
(x=4)						
10	90	385	37	0.083	65.4	1.02
(x=3.68)						
4.4	95.6	-234		No decomposition		
(x=21)						

**Conclusions**

The research shows that a relatively simple physico-chemical experiment provides a possibility for modeling an extreme reaction causing release of radionuclides from a storage facility, and to determine the energy characteristics of this reaction in the particular conditions under which it proceeds. By writing a plausible scenario of an accident at a radiochemical plant based on an analysis of indirect data and the energy characteristics of dangerous phenomena occurring as an accident develops, we can also pose and solve the reverse problem—establishing the possible consequences of an accident before it occurs, and in the ideal case, before a dangerous facility is placed into operation. In other words the design of a radiochemical plant (of its individual parts) must consider both predictable and unpredictable accidents, as is done with nuclear power stations.

Compiling a register of possible accidents at enterprises involved in the nuclear fuel cycle was attempted in [16]. Several scenarios of possible accidents were formulated and checked out in [17] for the radiochemical plant's department storing and processing highly radioactive liquid wastes and radioactive slurry.

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**Problems Regarding Lithium Batteries**

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[FBIS Translated Text]

**Summary**

The article deals with disposable and reusable lithium-type chemical current sources, basic problems of scientific nature (passivation and reversible action of the lithium electrode, intercalation of lithium ions into carbon materials, various kinds of anodes, etc.) being examined along with some recent results of scientific research on this subject.

**Introduction**

The appearance in the early nineteen seventies of a new type of chemical current sources, namely galvanic

cells with lithium in aprotic electrolyte solutions, was met with great interest by the users. Use of metallic lithium as the active cathode material has substantially increased the energy mass density (W.h)/kg or volume density (W.h)/l above that in conventional chemical current sources using various other electrochemical systems. Thus the energy content in Li/SOCl<sub>2</sub> (thionyl chloride) cells is about 600 (W.h)/kg or 900 W.h/l while that in Zn/HgO cells, having the highest energy content among all known cells, does not exceed 120 (W.h)/kg or 450 (W.h)/l. It is for this reason that lithium cells and batteries have in the past decade been extensively used as electric power supplies for small special-purpose devices as well as for small consumer appliances (watches, cameras, calculators, etc.). Multimillion-volume production of such cells has already been set up in many countries.

Still, the production volume of primary lithium cells amounts so far only to a small fraction of the total production of all disposable galvanic cells. The main causes preventing their broader application are:

- a) high cost (due to the necessity of using carefully purified reactants and due to still high production labor cost);
- b) apprehension as to absolute safety in use (as of now, lithium cells are not allowed in children's toys);
- c) limitations on the discharge power of lithium cells with hard cathode materials.

So far there has not yet been set up any large-scale production of high-quality reliable rechargeable lithium storage cells (reusable current sources).

Evidently, therefore, there still remain here (especially in the case of lithium storage cells) several unsolved problems: not only problems of engineering-technological nature and of economics but also problems of scientific nature. Many studies are being made in various countries for the purpose of solving them.

It is noteworthy that work on scientific problems in the field of lithium batteries not only helps solving application problems but also has started two new research trends in theoretical electrochemistry. The first one pertains to the mechanism of passivation of highly active metals (lithium) by electrolytic solutes in organic solvents and by strong inorganic oxidizers (sulfuryl chloride, sulfur dioxide, etc.). The specifics of the lithium passivation process are what makes production of lithium batteries feasible. The second trend pertains to the thermodynamics and the kinetics of electrochemical injection of lithium ions or anions into solid solutions (oxides, sulfides, carbon materials, etc.) by way of

solid-phase intercalation and, reversely, their extraction from such compounds by way of solid-phase decalation. Such processes take place at the anode in most lithium batteries and also at the cathode in some of them.

This report will briefly cover the aspects of scientific problems regarding lithium batteries and also some significant results of studies made on the subject in recent years.

#### Problems Regarding Disposable Lithium Batteries

Disposable lithium batteries can be divided into two groups: 1. cells with solid cathode materials such as oxidizers (MnO<sub>2</sub>, CuO, CF<sub>3</sub>, etc.) and with electrolyte solutes in organic solvents, 2. cells with liquid oxidizers acting also as solvent (thionyl chloride, liquefied sulfur dioxide, etc.). Each group has its specific features.

*Problem of local action in lithium cells (corrosion of lithium electrode).* This is a theoretical rather than practical problem, because a lithium electrode corrodes very little despite the strong thermodynamic irreversibility of lithium-electrolyte systems and especially of those where the electrolyte is a liquid oxidizer. The well known historic J.P. Gabano test<sup>1</sup> revealed that during 16 years of shelf time Li/CuO cells had lost less than 2 percent of their capacity. The data which indicate such a small loss of capacity correlate with microcalorimeter readings which indicate that corrosion of lithium and local action in these cells are attended by a release of only extremely little heat. This suggests that the basic requirements regarding necessary purity of solvents and other reactants (including maximum allowable traces of water) can be met in a manufacturing facility.

It has been known for a long time that a lithium electrode in contact with an organic solvent or thionyl chloride owes its high corrosion resistance to the presence of a special protective film on its surface. This film is still the subject of intent experimental and theoretical research. Among the many published studies on this subject ought to be mentioned the articles by D. Aurbach and associates<sup>2,4</sup> reporting in detail their research on the chemical composition and the physicochemical properties of this film in various solvents, also the work done by Ye.S. Nimon with others<sup>5,6</sup> on the mechanism of current conduction through this film and on the laws governing its growth.

Even though corrosion of lithium is very slow, it still poses several yet unsolved practical problems. Among them is, above all, the effect of interruptions of discharge on the further behavior of the cell. Numerous observations have shown that the corrosion rate is significantly higher immediately after than before a partial

discharge. The results of impedance measurements indicate that the resistance of the protective film on the lithium surface is much lower after discharge than initially. This problem is an extremely serious one when it comes to actual use of lithium cells, which are generally meant to operate in the intermittent-discharge mode. Unfortunately, observations made regarding this problem have not yet been sufficiently well organized.

**Problem of voltage dips after long shelf time.** Several years ago this was a very serious problem. The problem was that as a cell began to deliver current after a long shelf time, its voltage fell and remained for the first few seconds (or minutes) significantly lower than the normal discharge voltage. This phenomenon is still noticeable when any of the various kinds of lithium cells are used, but found to be most pronounced in the case of the Li/SOCl<sub>2</sub> system. It has to do with the fact that the resistance of the protective film on the surface of a lithium electrode increases drastically during the shelf time.

By now most lithium cell manufacturers have, it seems, satisfactorily solved this problem by adding to the solvent various agents (in most cases organochlorine compounds) which influence the rate of growth and the structurization of the protective film on the lithium surface.<sup>7</sup>

**Determining the internal state of cells as the indicator of ways to improve their reliability and safety.** The shelf life of disposable lithium cells is, as a rule, longer when they have not yet been discharged (3-5 years and sometimes even longer). For several applications it is necessary to verify that they will retain their full capacity after a long shelf time, i.e., that no random processes attended by excessive local action have taken place during those periods. Thus arises the problem of nondestructive inspection for monitoring the internal state of such cells. The problem is an important one in view of the definite possibility that individual cells may spontaneously explode, as is being periodically reported. Although these explosions occur very seldom (only several times a year), they do scare away many potential customers.

The only reliable way to monitor local action is to continuously or periodically determine the rate of heat release by a cell.<sup>8,9</sup> Because of the slowness of spontaneous discharge, this method requires the use of highly sensitive microcalorimeters and its procedure is very laborious. Many serious attempts were made to determine the internal state of cells by the method of electrochemical impedance spectroscopy.<sup>10,11</sup> Although the magnitude of the impedance during discharge has been found to somewhat depend on the residual capacitance of the

cell, this method does not yet yield sufficiently reliable and unambiguous data on the internal state of cells. The problem, therefore, is yet to be solved and work toward this end continues.

**Increasing the discharge power of cells with solid cathodic reactants.** Disposable lithium cells with a solid cathode material (fluoridated carbon CF<sub>x</sub>, copper oxide CuO, manganese dioxide MnO<sub>2</sub>, etc.) are potentially safer during operation than cells with liquid cathodic electrolytes (thionyl chloride SOCl<sub>2</sub>), but the maximum allowable discharge current density in them is much lower (not above 1 mA/cm<sup>2</sup>). Much work is now being done in search of new oxides, sulfides, and other compounds as electrode materials which will allow cell operation at high current densities.<sup>13,14</sup> The expectations are, however, very slim: the speed of electrochemical reaction is limited by the slowness of diffusion processes taking place in the solid phase, and to accelerate them enough is hardly possible.

A basic approach now taken by many lithium cell developers as a way out of this deadlock is to substantially increase the active surface area by making the electrodes very thin.<sup>15,16</sup> This is a sensible and relatively simple way to increase the discharge power of these cells. It requires great caution, however, because there is a danger that a cell with a large electrode surface area per unit cell volume may overheat as its thermal state changes and a "thermal runaway" may sometimes occur as a result.

#### Problems Regarding Reusable Lithium Batteries

Since sometime in 1991 the emphasis in research work on lithium batteries shifted toward development of reusable (storage) cells. The scope of their applications (radioelectronic equipment, video cameras, personal computers, etc.) is much broader than that for disposable ones. Besides, they also often solve a problem of mere economics: even for a limited number of charge-discharge cycles it costs much less to use rechargeable cells than relatively expensive disposable ones.

Despite the many optimistic forecasts, a large-scale production of lithium storage cells has still not been set up. This is due to the many not yet completely overcome difficulties and still unsolved problems.

**Cycling a lithium electrode.** Despite all efforts made during the past 10-15 years, there has not yet been developed a metallic lithium electrode suitable for cyclic duty. The sediment of metallic lithium deposited during the charging period has a very friable structure. It is fragile and its individual particles are surrounded by nonconducting reaction products so that, being thus encapsulated, they lose electrical contact with one another. As a result, the charge holding capacity of such

an electrode is underutilized and the cell life (in terms of cycles it can deliver) is shortened.

This peculiarity of a lithium electrode is, moreover, also attributable to the presence of a protective film on its surface, which makes the degree of electrode underutilization depend also on the structure and the properties of that film. The dependence of the life of a lithium electrode on the content of the electrolyte solution has been the subject of many studies.<sup>17-19</sup> Noteworthy among the important achievements in this area is the recently discovered powerful effect of adding CO<sub>2</sub> to the solution, which has been found to greatly improve the utilization of the charge holding capacity.<sup>20,21</sup> This effect is probably due to a change of the film composition: from oxide, hydroxide, and organolithium compounds before the addition of CO<sub>2</sub> to what is essentially lithium carbonate LiCO<sub>3</sub> in the presence of CO<sub>2</sub>.

As long as the problem of cycling a lithium electrode will not be completely solved, the cathode of a storage cell will have to contain a large surplus of metallic lithium (typically 4-10 times more than the amount required for a given stoichiometry) enabling it to recover the loss of charge holding capacity during each discharge-charge cycle.

Some time ago attempts were made to overcome this difficulty by using Li-Al alloys.<sup>22,23</sup> These alloys do partly contribute to a better utilization of the charge holding capacity, but they are not mechanically strong enough to withstand the drastic change of their specific volume from that in the charged state to that in the discharged state. This drawback can be remedied by using alloys of lithium with heavy metals such as alloys of the Wood type<sup>24</sup> but, owing to the low lithium content, the characteristics of such an electrode are on a per unit basis still poor. Much less research work on lithium alloys was later done during the past few years, this approach to development of a reversible lithium electrode evidently not being very promising.

*Carbon electrodes with intercalated lithium ions.* An auspicious alternative to aluminum alloys and heavy-metal alloys with lithium for intercalation of its ions was found to be carbon. Graphite containing interstitial lithium or another alkali metal (laminar graphite compounds) has been generally known for a long time.<sup>25</sup> In the mid-eighties there was published a report describing reversible electrochemical intercalation of lithium from its salts in aprotic solvents into carbon materials. A strong stimulus for further developments was provided by a report about commercial production of rechargeable batteries with a carbon cathode being set up in Japan.<sup>26</sup> As anode materials for such a battery source

had been selected compounds of oxides and interstitial lithium. The processes of charging and discharging such a storage cell thus become simply a transfer of lithium ions from anode to cathode and in the reverse direction respectively. Current sources of this type are called "rocking chair cells" or lithium-ion storage cells.

The essential favorable feature of carbon materials distinguishing them from aluminum and other metals is that intercalation of lithium does not significantly increase their specific volume. While the specific volume of Li-Al alloys is almost twice as large as that of pure aluminum, the specific volume of LiC<sub>6</sub> is only 10 percent larger than that of the original carbon material.<sup>27</sup>

This report by Japanese authors immediately caused a boom in research work on intercalation of lithium into carbon materials. While only one paper on this subject had been presented at the fifth International Conference on lithium batteries in Beijing (China) 1990, already 13 papers were presented at the sixth one in Muenster (Germany) 1992 and 33 papers at the seventh one in Boston 1994.

In the very first few publications was already demonstrated the possibility of long heavy cycling of carbon electrodes: up to thousands of cycles with up to 350 (mA.h)/g.<sup>28</sup> Just as important is the fact that electrodes with intercalation have a much higher explosion and fire resistance than metallic lithium electrodes. No highly active metallic lithium precipitates at such an electrode and no dendrites will form during the cathodic process, as long as it is not accompanied by recharge. Use of a cathode with intercalation thus also solves the problem of retaining contact within the charged active mass. It is for these reasons that research work in this direction has now been recognized as the most promising one toward development of rechargeable lithium current sources, despite the fact that the 372 (mA.h)/g theoretical capacity of LiC<sub>6</sub> is significantly lower than the 3828 (mA.h)/g one of metallic lithium. An advantage of a LiC<sub>6</sub> cathode is, moreover, that it does not require a surplus of lithium as does a metallic lithium cathode.

The basic problems regarding intercalation of lithium into carbon materials were defined in the very first few articles on this subject. It was established that both the degree of intercalation and the activity (i.e., equilibrium potential) of a lithium intercalation depend strongly on the nature of the carbon material, especially on its crystallinity or degree of graphitization. The truth is that the data reported by various authors are extremely inconsistent. According to some data<sup>29</sup>, for instance, reversible intercalation was possible only into materials with a well defined crystalline graphite structure and impossible into amorphous materials, while according

to other data<sup>29</sup> only intercalation into an amorphous material had been possible.

An analysis of the various publications indicates that the best materials for reversible intercalation of lithium are amorphous ones containing "mesophase" (seeds of graphite crystallites) in their structure. Such materials are coke, pyrographite, and materials produced by pyrolysis (carbonization) of polymers. This conclusion is very well verified by the results of studies made concerning intercalation of lithium ions into products obtained by pyrolysis of the same compound at various temperatures.<sup>30,31</sup> These studies have revealed that the optimum temperatures for pyrolysis of such compounds happen to be exactly the temperatures at which it will yield the desired electrode materials and that these temperatures lie within the 2000-2500°C range.

Intercalation of lithium into a well defined graphite structure can, as a rule, yield the  $\text{LiC}_6$  compound (i.e., in  $\text{Li}_x\text{C}_6$ ). The curve depicting the dependence of the  $\text{Li}_x\text{C}_6$  potential on the degree of intercalation (x) usually includes a series of almost horizontal steps corresponding successively to the  $\text{LiC}_6$ ,  $\text{LiC}_{12}$ ,  $\text{LiC}_{18}$ , ... compounds.<sup>32</sup> It is to be noted that these compounds by themselves are known, having been isolated and well identified. Intercalation of lithium into graphite structures takes place at potentials ranging from 0.4 V to zero relative to the lithium electrode potential, the activity of intercalated lithium then being sufficiently high.

Meanwhile, intercalation of lithium into amorphous and mesophase structures takes place at potentials ranging from 0.9 V to zero and the dependence of the electric potential on the degree of intercalation (x) follows a smooth curve, which characterizes a homogeneous system.

The correlation between intercalation of lithium into carbon materials and the characteristics of their crystalline structure has not yet been on the whole conclusively established and, therefore, still remains on the list of important ones in this area to be dealt with.

Problems still before researchers working on intercalation of lithium into carbon materials fall into two categories: 1. problems common to all intercalation compounds, 2. problems which relate only to intercalation into carbon materials. Those in the first category include: a) thermodynamics of the intercalation process (absolute magnitude of the lithium activity in the intercalation compound, its dependence on the composition x of that compound and on the temperature), b) kinetics of diffusion (or other modes of transfer) of lithium ions in the solid phase, c) cointercalation of the solvent and

other ions from the electrolyte solution, d) kinetics of lithium injection into the solid phase.

Problems of the second category have to do with the surface of carbon materials, namely its activity and catalytic properties. It was already established in the first few studies that this surface (especially the surface of strongly graphitized materials) catalyzes reduction of organic solvents.<sup>33,34</sup> During the first cathodic polarization (charging) a large portion (20-40 percent) of the charging current is used up not for intercalation of lithium but for reduction of the surface carbon groups and mainly of the electrolyte components (solvent, anion). The products of this reduction reaction form a thin film on the electrode surface which inhibits further reduction of the electrolyte, its action being analogous to that of the protective film on the lithium surface. Because of this, during subsequent cycling the entire charging current will already be used up only for intercalation of lithium. The properties and the composition of the film on the surface of carbon materials have also been thoroughly studied.<sup>35,36</sup>

All the phenomena described here depend quite strongly on the content of the electrolyte solution, particularly on the nature of the solvent.<sup>36</sup>

*Problems regarding the anode of rechargeable lithium batteries.* In the first few versions of a reusable current source, with a metallic lithium cathode, as anode materials were used chalcogenides or oxides of variable-valence metals receptive to reversible intrusion of lithium.<sup>37</sup> Most popular were titanium sulfide  $\text{TiS}$  and molybdenum disulfide  $\text{MoS}_2$ , also various vanadium oxides. A major drawback of sulfide electrodes is that injection of lithium into them takes place at relatively low potentials (1.5-2.5 V relative to the lithium potential) and the discharge voltage therefore is low. Use of vanadium oxides or manganese oxides made it possible to raise the nominal cell voltage to 3 V. Meanwhile the replacement of metallic lithium in the cathode with lithium intercalations and particularly its intercalation in carbon, where its operating range of potentials is about 0.5 V more positive than that of metallic lithium, has been a forward step in the development of electrodes operating at still more positive potentials. Materials for electrodes operating at a potential of about 4 V were found among complex oxides with lithium: lamellar lithium-cobalt oxides,<sup>26</sup> lamellar lithium-nickel oxides,<sup>38</sup> and lithium-manganese spinels.<sup>39</sup> In fact, most of the studies made in recent years on anodes for rechargeable lithium batteries dealt with just these compounds. It was established first of all that under the proper conditions these oxides will withstand long cycling without degradation of their characteristics.

One of the main problems with producing cobalt oxide, nickel oxide, and manganese oxide anodes is the problem of a sufficiently reliable technology which will yield materials receptive to intercalation of lithium. While a  $\text{Li}_x\text{CoO}_2$  compound can be obtained by simple solid-phase interaction of lithium and cobalt carbonates, the technology of producing  $\text{Li}_x\text{NiO}_2$  compounds or  $\text{Li}_x\text{Mn}_2\text{O}_4$  compounds (spinel) is much more intricate and includes, among others, a solid-phase reaction of  $\text{LiOH}$  with  $\text{NiO}_2$  or with  $\text{MnO}_2$  respectively. An essential factor here is that the properties of the Li-Mn oxides depend strongly on their stoichiometry. Although these relations have been studied,<sup>40</sup> the problem of optimizing the stoichiometry of high-voltage oxides still remains on the list of important ones to be dealt with.

The main advantage of these oxides, namely their high electric potential during intercalation of lithium (i.e., during discharge), is also their main drawback: strong oxidizing action on the solvent. Oxidation of the solvent during charging raises the internal pressure in the cell, increases the consumption of active substances, and in an open circuit (especially at higher temperatures) causes local action at the anode. There thus arises the problem of selecting an electrolyte which has a high oxidation resistance. It is just possible that a solution to this problem will be found in using solid polymer electrolytes.

The oxidizing activity of oxides depends on their nature and local action is, for instance, stronger at  $\text{Li}_x\text{CoO}_2$  electrodes than at  $\text{Li}_x\text{NiO}_2$  electrodes.<sup>41</sup> Corrosion of  $\text{Li}_x\text{Mn}_2\text{O}_4$  spinels slows down after they have been doped with cobalt<sup>42</sup> or other metals.<sup>43</sup> Many researchers believe that the characteristics of mixed oxides may be better than those of the simple oxides.<sup>44,45</sup>

Although anodes for rechargeable lithium batteries have been the object of many studies, the main fundamental problems regarding their constitution have by far not yet been solved. There are no definitive data available regarding the dependence of their open-circuit (or equilibrium) potential on the temperature and on the degree of lithium intercalation in various electrolytes, no data at all regarding the kinetics of solid-phase diffusion. There still remain open questions concerning the surface properties of such materials and the related problem of the contact resistance between particles of the active material. Just as pressing is the problem of degradation of the active material during cycling, this problem being common to all types of secondary chemical current sources.

*Use of conducting polymers as anode materials.* In the second half of the nineteen eighties much work was done on the use of conducting polymers (polyacetylene, polypyrene, polythiophene, etc.) for the anode of lithium

batteries. These polymers are receptive to reversible intercalation of anions at potentials 2-3 V more positive than that of the lithium electrode. At some lowest degree of intercalation (doping level) they become n-type conductors adequate for serving as electrode materials.

Despite the considerable effort invested in it, this concept has not been pursued to the point of practical realization. There were several reasons. The polymers turned out not to be chemically stable enough during operation of such a rechargeable cell, thus shortening its life. The degree of anion intercalation was low ( $x=0.1-0.15$  per CH group of the polymer). The main reason was, however, the very intercalation of anions rather than cations. During charging lithium ions would discharge (or move into the carbon matrix) at the cathode and anions would move into the polymer at the anode, the dissolved salt thus being used up. A surplus volume of electrolyte is therefore required, which appreciably decreases the specific capacity of such a battery. Work on this project has been now drastically curtailed.

*Rechargeable lithium batteries with solid polymer electrolyte.* In recent years there started a new trend in the construction of rechargeable lithium batteries: reversibly operating cells with a solid polymer electrolyte instead of a liquid solution. Use of a solid electrolyte serves two main purposes: avoids outflow of liquid in every spatial arrangement (or under mechanical overload) and makes it safe to use very thin cells with a large electrode surface area per unit volume, i.e., to appreciably increase the discharge power per unit mass of a battery.

In the first attempts the solid electrolytes were complexes consisting of polyethylene oxide (PEO) and a lithium salt as, for example, the (PEO)<sub>n</sub> $\text{LiClO}_4$  complex. This complex has a sufficiently high electrical conductivity at elevated temperatures:  $\sigma=10 \mu\text{S}/\text{cm}$  at 100°C. At lower temperatures such a solid electrolyte undergoes transition into amorphous and crystalline states (vitrification), with its electrical conductivity decreasing sharply to below the minimum allowable. In view of this, the main efforts in here were focused on attempts to increase the electrical conductivity of solid electrolytes at still lower temperatures as, for instance, room temperature.

In several studies other polymer compounds were used as solid electrolytes. Some encouragement was expected from a poly[bis(methoxyethoxy)phosphasene] (MEEP) polymer. Such an electrolyte has a higher electrical conductivity at room temperature, but tests have revealed that it makes poor contact with cathode materials.<sup>46</sup> As another possible variant of a solid polymer electrolyte material was considered polyvinylidene flu-

oxide.<sup>47</sup> However, the electrical conductivity of all these realizable polymer electrolytes is still not high enough for an acceptable battery performance at room and lower temperatures.

A different approach has been found to be more promising: use of polymers plasticized by addition of lithium salts in organic solvents whose dielectric permittivity is high and vapor pressure is low. The drawback of such materials is the appreciably lower mechanical strength of the solid electrolyte film. In one report<sup>48</sup> the authors proposed new variants of polymer electrolyte materials: polyacryl nitride and polyvinyl chloride plasticized by solutions of lithium salts in ethylene carbonate. The electrical conductivity of such electrolytes reaches 2 mS/cm at 20°C and 0.2 mS/cm at -40°C. These figures are very encouraging.

### Conclusion

The basic problems regarding disposable lithium batteries (local action at the lithium electrode and in the whole cell, voltage dip, and others) have been solved to the extent necessary for setting up relatively large-scale production of such batteries. There nevertheless still remain open some individual questions concerning, first of all, assurance of their absolutely safe operation. Another rather serious problem is lowering their production cost. This can be done not only by improvement of the fabrication process but also by use of less expensive new electrode materials, i.e., new electrochemical systems. Concerning this possibility, certainly very interesting are the studies made on use of natural or synthetic pyrite as the active material in disposable lithium batteries.<sup>49</sup> Important in a number of applications is also the feasibility of raising the specific power by constructing, for instance thin-film cells.

At the same time development of rechargeable lithium batteries still requires a solution of many problems of scientific nature before mass production can be set up. These problems pertain essentially to assurance of a sufficiently long life (in terms of charge-discharge cycles). The target of most development work in this area are storage batteries having a specific energy content of 150-200 (W.h)/kg and a life expectancy of several hundred (up to a thousand) charge-discharge cycles with assurance of absolutely safe operation. There also arises the problem of producing relatively large units, chiefly for the electric power system of electric cars. Many researchers in various countries, including Russia, are at this time working on scientific and technological problems to this.

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**Third Conference of CIS Countries on Lithium Batteries**

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[Report by N.V. Korovin and A.M. Skundin, Institute of Electrochemistry imeni A.N. Frumkin at Russian Academy of Sciences, Moscow; UDC 541.136.1]

[FBIS Translated Text] Because of their high specific energy content along with good performance and long shelf life at low temperatures, lithium batteries are being more and more extensively used in various branches of technology. Worldwide production of primary lithium batteries has reached a volume of several million annually. Rechargeable (secondary) ones are put on the market in small quantities. Lithium batteries are extensively studied in all developed countries of the world. Scientific research and experimental engineering work on lithium batteries is also being done in many laboratories, institutes, and enterprises in Russia and other CIS countries. Conferences on problems of lithium batteries are, therefore, very important.

At the Third Conference of CIS countries on lithium batteries took place in Yekaterinburg on 4-7 October 1994. One can say that such conferences have already become a tradition. The first (All-Union) one was held in Novocherkassk in September 1990 and the second one was held in Saratov in September 1992. Despite all the problems of logistics and economics, the conference in Yekaterinburg was attended by 84 participants from 21 cities in Russia, Ukraine, and Belarus. They represented 36 organizations, enterprises, and companies engaged in development and production of lithium batteries. Representatives from Georgia and Kazakhstan were, unfortunately, not able to attend.

The Conference covered all problems regarding lithium batteries, ranging from basic research on electrochemical and physicochemical processes to problems of their production and specific applications. The Conference was organized into plenary lectures and follow-up sessions. Doing away with the traditional section meetings (the various sections working "in parallel" as they did, for instance, at the First Conference in Novocherkassk) was again found to be entirely justified.

Special attention at the plenary sessions was paid to general problems regarding development of lithium batteries. Particularly noteworthy were here N.V. Korovin's report on "Development of Chemical Current Sources and Role of Lithium-Type Chemical Current Sources", and V.S. Bagotskiy's on "Problems of Scientific Nature Regarding Lithium Batteries."

The proceedings at this Third Conference revealed that, despite certain difficulties associated with scientific research and engineering development work, some definite successes had been achieved during the two years since the Second Conference on Lithium Batteries. Much interest was aroused by the following lectures: O.N. Yefimov's "Solid Polymer Electrolytes in Lithium Batteries", A.M. Skundin's "Reversible Intercalation of Lithium into Carbon Materials", Ye.I. Burmakin's "Modern Trends in Development of Lithium-Transporting Solid Electrolytes", I.A. Kedrinskiy's "Behavior of Lithium in Chemical Current Sources", A.A. Andriyko's "Dynamic Instability in Lithium and Protective Film Systems", N.N. Batalov's "Research on Design of High-Temperature Storage Batteries", V.K. Kulifeev's "Utilization of Scrapped Lithium Batteries and of Tailings from Their Production Process."

The program of the follow-up session listed 74 lectures covering five topics: "Primary Current Sources and Storage Batteries", "Lithium Electrode", "Cathode Materials", "Electrolytes", and "High-Temperature Cells." In addition, the program of the Conference included oral and promotional presentations, most of them (just as the follow-up lectures on "Primary Current Sources and Storage Batteries") dealt with concrete developments and with organizing the production of lithium batteries. The resolution adopted by this Conference includes a special mention of the fact that the number of organizations working on and producing lithium batteries along with the production volume and the assortment of chemical current sources have all substantially increased during the time since the Second Conference. In connection with this growth there ought to be mentioned particularly TOO "Raduga" [Rainbow] in Podolsk, the TOO "Sulfid" [Sulfide] in Krasnoyarsk, the Novosibirsk Chemical Concentrates Plant, the Angarsk Electrochemical Combine, the Scientific Research and Manufacturing Engineering Office (NKTB) for Chemical Current Sources in Novocherkassk, the AO Scientific Research Institute of Chemical Current Sources in Saratov, also the already set up production of aprotic electrolytes in Tomsk, Slavgorod, Krasnoyarsk, Kiev and Dnepropetrovsk, the production of lithium foil in Saratov, and the production of carbon polyfluoride in Angarsk. The annual volume of lithium battery production in CIS countries is of the order of tens of millions. The production capacity, the one already in place plus the one to be installed, will be able to meet the demand for primary lithium batteries on the CIS market.

At the Conference was, moreover, recognized the noticeable lag behind the scientific research and experimental engineering work at the global level, not to mention the lag behind global production of secondary

lithium batteries. Unfortunately, owing to the lack of financial support, work on the utilization of scrapped lithium batteries and on other ecological problems has been practically halted. The Conference has taken note of the lower level of information exchange within the CIS not only concerning the status of work on lithium batteries in its member countries but also on the overall global status of work in this field.

It was resolved at the Conference that an initiative be taken toward establishment of foundation promoting cooperation in the development of chemical current sources. Such a foundation could provide financial support for conducting conferences, organizing exhibitions, etc.

On the whole, the Third Conference on Lithium Batteries was quite well organized, thanks to the tremendous effort made for its preparation by the staff of the Institute of High-Temperature Electrochemistry in the Ural Department of the Russian Academy of Sciences.

The next Fourth Conference on Lithium Batteries is to take place in 1996 in Chernogolovka, Moscow Oblast, and to be ranked as a scientific one. Organization of that conference has been assigned to the Institute of Chemical Physics and Electrochemistry at the Russian Academy of Sciences.

#### **Effect of Steady Illumination on Properties of 'Lithium - Anhydrous Solution' Interface Anhydrous Solution**

957A0702A Moscow ELEKTROKHIMIYA in Russian  
Apr 95 Vol 31 No 4, pp 350-354

[Article by Ye.S. Nimon, A.V. Shirokov, N.P. Kovynov, and A.L. Lvov, Saratov State University; UDC 541.138-145.4 (manuscript received 31 May 94)]

[FBIS Abstract] An experimental study of the response of a "lithium - passivating film - anhydrous electrolyte solution" system to steady illumination was made in search of answers concerning the nature of attendant processes. Tests were performed with three electrolytes: (1) 1 M LiClO<sub>4</sub> in propylene carbonate, (2) 1 M LiClO<sub>4</sub> in propylene carbonate + dimethoxyethane = 1:1 mixture, (3) 1 M LiClO<sub>4</sub> in thionyl chloride. The water content in each solution did not exceed 0.005 wt.%. As the source of 300-600 nm visible and near-ultraviolet light was used a superhigh-pressure mercury lamp with focusing quartz optics and with a water filter for cutting off infrared light. A xenon lamp was used for momentary illumination of the electrode with flashes of 40 J energy and about 100  $\mu$ s duration. The transmission spectra of 1 mm thick electrolyte layers were recorded with a Specord M40 spectrophotometer. Photoelectrochemical measurements

were made in hermetic cells with a third electrode and with a optical quartz window. In this way were recorded potentiodynamic curves in darkness and in steady light (10-1800 h). An analysis of the data has revealed some characteristics of passivating films on lithium, namely dependence of their resistance to ion current and of the lithium electrode potential on the length of illumination time (0-1600 s lamp-on time followed by afterglow time). Measurements made at temperatures over the 273-343 K range have yielded straight  $\log \sigma_0 T$  vs.  $1/T$  curves characterizing the temperature dependence of the electrical conductivity of the passivating films, in darkness and in light. Also the current transients were measured during illumination by light pulses with short rise time and long fall time, the current waveform found to be almost duplicating the light pulse waveform. On the whole the results reveal a strong thermal effect of steady illumination on the electrode, the passivating film and the adjacent electrolyte layer. Illumination by flashes induced a photocurrent consisting of a "fast" component flowing to the cathode and a "slow" component due to heating. The latter component could be suppressed by illumination with still powerful but much shorter flashes. Figures 5; references 23.

#### **Study of Solid Electrolyte Layers in I<sub>2</sub>P2VP/Li Batteries by Galvanostatic Pulse Method**

957A0702B Moscow ELEKTROKHIMIYA in Russian  
Apr 95 Vol 31 No 4, pp 355-358

[Article by Ye.S. Nimon, A.V. Shirokov, N.P. Kovynov, and A.L. Lvov, Saratov State University; UDC 541.138-145.4 (manuscript received 31 May 94)]

[FBIS Abstract] An experimental study was made concerning the properties of solid electrolyte layers in hermetic iodine-lithium batteries with a [I<sub>2</sub>][poly-2-vinyl pyridine (P2VP)]-complex cathode material and a metallic lithium anode material. Cells were assembled in a dry argon atmosphere, the cathode mass with a 1.54 cm<sup>2</sup> active surface area having been pressed into a fluoroplastic holder. Such cells, with a cathode-limited capacity of about 60 mA.h, were placed in a stainless steel housing and the latter sealed with a compound. All measurements were made in a thermostat at a 298 K temperature. A generator of square voltage pulses convertible into current pulses was used for measuring the resistance and the differential capacitance of these cells. A pulsed potentiostat was used for recording their current-voltage characteristics. The results indicate that the solid electrolyte layer, which forms when cathode and anode are brought into direct contact, acts as their separator and thus becomes the dominant factor influencing the electrochemical behavior of such batteries. The current-voltage characteristics fit the model of space-charge lim-

itation, but with the total current also including the contribution of intrinsic current carriers. Measurements of the transport characteristics have revealed a relaxation effect: a decrease of the Li<sup>+</sup> ion mobility followed by its very slight increase, a monotonic decrease of the Li<sup>+</sup> ion conductivity, and an increase of the Li<sup>+</sup> ion concentration over a period of up to 300 h. Figures 7; references 9.

**State of Surface Layers on Lithium in Anhydrous Media Containing Modifier**

957A0702C Moscow ELEKTROKHIMIYA in Russian Apr 95 Vol 31 No 4, (manuscript received 10 Feb 94, after revision 15 Jun 94) pp 359-364

[Article by V.M. Ovsyannikov, A.G. Demakhan, A.G. Zhukov, and V.M. Zhivaykin, Scientific Research Institute of Chemistry at Saratov State University; UDC 541.135.5:539.216.2]

[FBIS Abstract] The surface of a lithium electrode was examined after it has made contact with various electrolyte solutions based on aprotic solvents and containing various modifiers. The working electrode was the tip of lithium rod with a 5-10 mm<sup>2</sup> large geometrical surface pressed into a glass or teflon holder. A reference electrode and an auxiliary electrode were included in the cell. The electrolyte was 1 M LiClO<sub>4</sub> solution in propylene carbonate with either methyl acetate or triethylamine or both added as modifier. Grade "pure" triethylamine had been held over solid KOH for 7-10 days and then distilled, whereupon its middle fraction was dried with molecular grade NaA sieves and then again distilled. It was added to the electrolyte solution in amounts corresponding successively to 0.05/20 - 0.05/10 - 0.1/10 - 0.75/10 wt.%/ml concentrations. Methyl acetate was added in an amount corresponding to 25 wt.% concentration. Electrical measurements were made by the galvanostatic pulse method at a 298 K temperature, current pulses of 50  $\mu$ s duration having amplitudes of 2.0-3.0 mA/m<sup>2</sup> being applied to the main electrode. The differential capacitance of such a lithium electrode was calculated from the slope of the initial segment of its potential vs. time curve representing the electrochemical process kinetics and then used, in the formula for a parallel-plate capacitor, for calculating the thickness of the passivating layer on the lithium surface. The measurements have yielded the dependence of the polarizational film resistance on the length of lithium-solution contact time. Mass spectrometry has yielded the dependence of the intensity of secondary ion (CH<sub>3</sub><sup>+</sup>, LiO<sup>+</sup>, Li<sub>2</sub>Cl<sup>+</sup>) currents on length of film sputtering over the lithium surface, no peaks attributable Li:N<sup>+</sup>, Li:NH<sup>+</sup>, Li:NCH<sub>3</sub><sup>+</sup> ions having been detected. Measurements were supplemented with mi-

crostructural examinations of the lithium surface after it come in contact with the electrolytic solutions. Figures 3; tables 1; references 9.

**Corrosion of Anode in Lithium Batteries**

957A0702D Moscow ELEKTROKHIMIYA in Russian Apr 95 Vol 31 No 4, pp 365-372

[Article by I.A. Kedrinskiy, L.K. Gerasimova, V.I. Shilkin, and I.I. Shmydko, Scientific-Industrial Complex "Sulfid" at Siberian Institute of Technology, Krasnoyarsk; UDC 620.193.23 (manuscript received 26 Apr 94)]

[FBIS Abstract] Corrosion of the anode in lithium batteries during storage is analyzed theoretically, considering that after such a battery has been hermetically sealed in a container it becomes a systems essentially off equilibrium and that then chemical and physical processes tend to restore the equilibrium during subsequent storage. A chemical reaction of lithium with any particular oxidizer produces a nonstoichiometric ionic compounds which then forms an insulating layer on the lithium surface. Although replacement of a gaseous medium with an anhydrous liquid electrolyte does not fundamentally alter the functioning of the galvanic cell responsible here for corrosion of lithium, formation of that insulating interface at the boundary between lithium and the liquid electrolyte boundary is being investigated with emphasis on the high chemical activity of metallic lithium. Because of this high chemical activity, that insulating layer remains indestructibly continuous, any "break" being immediately "cured", and thus permanently separates metallic lithium from the ambient medium. It is accordingly a multifunctional insulating layer and as such necessarily participates in the electrochemical process attending operation of such that galvanic cell while the Li  $\rightarrow$  Li<sup>+</sup> + e<sup>-</sup> reaction takes place at the lithium (electrode of the first kind) surface. Most experiments were performed with 220-30 mm long lithium wire 0.3 mm in diameter. The cell contained also a reference electrode and two pipelets for intake of gas or vapor and liquid electrolyte respectively. The conditions of the lithium surface depended on the atmosphere (inorganic or organic medium) which the electrode had been extruded: molecular oxygen or SOCl<sub>2</sub> vapor; diethyl ether (C<sub>2</sub>H<sub>5</sub>)<sub>2</sub>O, acetonitrile CH<sub>3</sub>CN, dimethyl formamide (CH<sub>3</sub>)<sub>2</sub>HCON, or other organic vapor; liquid with the same solvents and with various doses of one or another additive. The corrosion rate was measured by both the gravimetric method and the Oldham-Mansfield electrochemical method. The results of comprehensive tests validate the the use lithium electrode in anhydrous electrolytes and indicate an exact agreement with the Nernst equation. They depict the kinetics of the lithium

electrode potential and its dependence on the water content in the electrolyte when an insulating layer is being formed on the lithium surface either by a nonstoichiometric compound alone or by a such a compound with Fe, Ti impurities coming from an electrolyte which had absorbed them following corrosion of the cell container made of a Ni-Cr-Ti steel. The concentration of metal ions in the electrolyte was measured by the photocolorimetric method and its water content was measured using an automatic coulometric titration apparatus with spent Fischer's reagent. The data indicate that long shelf life of a lithium electrode can be assured by maintaining the total initial water content in the electrolyte below 0.01 vol.% and by not letting the total concentration of ions of polyvalent metal ions in it exceed 1.0 mg/ml throughout the entire storage period. Figures 8; tables 4; references 16.

#### Activity of Lithium Intercalated into Carbon Materials

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Apr 95 Vol 31 No 4, pp 373-375

[Article by A.M. Skundin and O.Yu. Yegorkina, Institute of Electrochemistry imeni A.N. Frumkin at Russian Academy of Sciences, Moscow; UDC 541.138.2 (manuscript received 28 Apr 94)]

[FBIS Abstract] Equilibrium potentials of carbon cloth electrodes were measured during intercalation of lithium from  $\text{LiClO}_4$  solutions in various organic solvents (propylene carbonate  $\text{C}_4\text{H}_6\text{CO}_2$ , propylene carbonate + 70% dimethoxyethane mixture,  $\gamma$ -butyrolactone  $\text{C}_2\text{H}_6\text{O}_2$ ). Measurements were made by the quasi-steady galvanostatic method, with one carbon cloth electrode wrapped in a solid polypropylene separator jacket together with two auxiliary lithium electrodes and an unpolarized lithium reference electrode in each fluoroplastic container. Prior to the cell assembly, all electrodes had been sintered for two hours under vacuum at a  $180^\circ\text{C}$  temperature and then placed in a box containing argon with an only 50-65 ppm moisture content. A structural and phase analysis in a DRON-3 x-ray diffractometer revealed hardly any graphite content in the cloth. Cathodic polarization curves were plotted with a current of 0.1 mA magnitude (0.33 mA/g). The difference between cathodic and anodic polarization curves did not exceed 3 mV, indicating a near-equilibrium state typical of not at all or only slightly graphitized materials. The dependence of the equilibrium potential on the degree of intercalation, i.e., on the lithium content  $x$  in  $\text{Li}_x\text{C}_6$  was found to fit not the Nernst equation with a constant activity coefficient but rather a power law where the exponent  $n$  associated with the activity coefficient

depends on the electrolyte material. Figures 3; tables 1; references 8.

#### Diagnostic Testing of $\text{Li}/\text{SOCl}_2$ Cells by Impedance Method

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Apr 95 Vol 31 No 4, pp 375-382

[Article by L.S. Kanevskiy, V.S. Bagotskiy, and Ye.A. Nizhnikovskiy, Institute of Electrochemistry imeni A.N. Frumkin at Russian Academy of Sciences, Moscow; UDC 541.138 (manuscript received 20 Jun 94)]

[FBIS Abstract] Determining the condition of  $\text{Li}/\text{SOCl}_2$  cells by the impedance method is considered, correlations being sought between changes in their condition and changes in their electrical impedance. Relevant tests were performed with five cells having different shapes and size having a nominal capacity of 0.17 A.h (TL-53), 0.6 A.h, 1.2 A.h, 4.5 A.h, 1.5 A.h (TL-85) respectively. Impedance measurements were made in the potentiostatic mode with a Solartron 1255 frequency analyzer covering the 0.1 Hz to 100 kHz range and using a Solartron 1286 electrochemical interface at a  $20(+/-)2^\circ\text{C}$  temperature. Both a counter-e.m.f. equal to the cell voltage and a 10 mV analyzer signal were applied to such a cell. These measurements have yielded X-R hodographs (X- capacitive reactance, R- resistance) of such a cell before and during gradual discharge. Both the d.c. (maximum) resistance and the maximum reactance (at some critical frequency) increased, as thus also did the initial hodograph slope (at the  $X=0$  and  $R=0$  or later  $R > 0$  point, as discharge of the cell continued. A special experiment was performed for the purpose of verifying the hypothesis that passivation of the lithium surface upon rupture of the continuous surface film by a current pulse would not alter the cell condition. The results of this experiment do, indeed, indicate that following a momentary discharge the impedance slightly dips but then promptly returns to its initial level or even exceeds it. Another confirmation of that hypothesis are the results of reactance measurements made after a momentary discharge and over a subsequent 17 months long storage time. In two other experiments was measured the dependence of that maximum capacitive reactance on the length of time taken for complete depletion of a cell and on the length of storage time respectively. No direct correlation has been established between the impedance of  $\text{Li}/\text{SOCl}_2$  cells and their depletion level, except that their impedance is much lower after partial discharge or local action. Two technological causes of the lack of correlation are: 1. impossibility of precisely reproducing precisely the same surface finish (degree of oxidation and roughness level; 2. inevitable inequality of electrolyte volumes, especially when due to wide

variation of the  $\text{SO}_2$  concentration. Figures 7; tables 3; references 18.

**Effect of Catalysts on Passivation of Lithium in Thionyl Chloride Electrolyte**

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[Article by L.S. Kanevskiy, M.B. Avdalyan, and T.L. Kulova, Institute of Electrochemistry imeni A.N. Frumkin at Russian Academy of Sciences, Moscow; UDC 541.138:3 (manuscript received 20 Jun 94)]

[FBIS Abstract] An experimental study of  $\text{Li}/\text{SOCl}_2$  cells was made concerning the effect of organic catalysts added for the purpose of raising the voltage as well as the already high energy content. Electrode pairs, a lithium plate and a carbon black plate with a glass separator between them, were placed in fluoroplastic containers with electrolyte. The electrolyte was  $\text{LiAlCl}_4$ , obtained by successively dissolving dry twice recrystallized  $\text{LiCl}$  and high-purity  $\text{AlCl}_3$  in twice distilled  $\text{SOCl}_2$ . All electrodes were produced and all cells were assembled in an argon atmosphere. Impedance measurements including the Warburg resistance were made in the potentiostatic mode with a Solartron 1255 frequency analyzer covering the 0.1 Hz -100 kHz range and using a Solartron 1256 electrochemical interface. As catalysts were added macrocycle compounds, specifically porphyrins. Measurements were made after storage periods ranging from 1 h to 3370 h and during subsequent discharge. For comparison, also cells with platinum catalyst and without catalyst were similarly tested. The results reveal that these organic catalysts perform better than platinum by not contributing to corrosion of the lithium anode and by thus not causing a large initial voltage dip as the cell begins to discharge. An analysis of the data, with the passivating film replaced by its equivalent electric circuit, indicates furthermore that the presence of such a catalyst alters the electrical properties of the passivating film so as to aid the cathodic process and to retard polarization. Figures 5; tables 2; references 16.

**Synthesis and Study of New Composite Solid Electrolytes Based on Polyethylene Oxide with Aluminum Oxide and Crown Ether**

957A0702H Moscow ELEKTROKHIMIYA in Russian Apr 95 Vol 31 No 4, pp 388-393

[Article by O.V. Yarmolenko, A.Ye. Ukshe, T.I. Movchan, O.N. Yefimov, and A.F. Zuyeva [deceased], Institute of Chemical Physics at Russian Academy of

Sciences, Chernogolovka; UDC 541.133.08 (manuscript received 21 Mar 94)]

[FBIS Abstract] New composite solid electrolytes with PEO polymer (polyethylene oxide) base were synthesized and studied, those synthesized earlier by formation of polymer and metal salt complexes not being quite satisfactory. The  $(\text{PEO})_4\text{LiClO}_4$ -complex material was synthesized by dissolving first ceramic  $\text{LiClO}_4$  and then dry PEO polymer in pure acetonitrile at a 40-50°C temperature, the ceramic having been first roasted under vacuum with the temperature stepwise raised to 210°C. Their weight fractions 79.30% PEO and 20.70%  $\text{LiClO}_4$  were selected on the basis of having eight oxygen atoms per lithium ion. To this basic material were added fillers: 1. 10 wt.%  $\beta\text{-LiAlO}_2$  or  $\gamma\text{-LiAlO}_2$  produced by synthesis of  $\text{Al}_2\text{O}_3$  and  $\text{LiOH}$  in a muffle furnace at 600°C or 800°C respectively ( $\alpha\text{-Al}_2\text{O}_3$  powder of the 1  $\mu\text{m}$  size fraction having been first roasted at temperatures up to 400°C), 2. 2,4-dioxo-16-crown-5 (1,5,8,11,14-pentaoxacyclohexadecyl-2,4-dione) ether melting at 66.5-68°C. Three of these composites contained 9.09 wt.%  $[\text{Al}_2\text{O}_3]$ , 37.20 wt.% [crown ether], and 9.08 wt.%  $[\text{Al}_2\text{O}_3]$  + 33.82 wt.% [crown ether] respectively. The solutions were deposited on teflon substrates for slow drying in an argon atmosphere until they formed solid films. The films were then kept in vacuum flask 2-4 h at a 40°C temperature for removal of residual traces of the solvent. The cathodes for these electrolytes were made of thermally structurized polyacrylonitrile. The experimental procedure included: 1. measuring the thickness of films with a standard micrometer; 2. phase analysis of  $\text{LiClO}_4$  ceramic in a DRON-3 diffractometer; 3. impedance spectroscopy of the electrochemical cells on an impedance bridge, covering the 0.1 Hz - 200 kHz frequency range and using an a.c. test signal with an amplitude of about 3 mV without a d.c. bias. Symmetrical cells with stainless-steel blocking electrodes and asymmetrical cells with one lithium counterelectrode each were thus tested. Charge was injected into cells while measurements were made with a potentiostat and a coulometer, the latter including a voltammeter and a digital counter. An analysis of the results, with a cell represented by its equivalent electric circuit including the Warburg resistance, reveals the effect of inert additives in the polymer on the impedance hodograph of a cell, both  $\text{Al}_2\text{O}_3$  and crown ether lowering the volume resistance substantially. While inert  $\text{Al}_2\text{O}_3$  does not change the form of the hodograph and thus the relaxation characteristics of the steel/polymer interface, active-donor crown ether substantially lowers the admittance of the electrode interface and thus alters the character of electrode relaxation. Contact between modified-PEO electrolytes and a metallic lithium electrode was surprisingly found to be stable and to almost

insignificantly increase the admittance of these electrolytes. Addition of crown ether evidently improves the characteristics of polyethylene oxide, because crown-ether-Li complexes are formed which may weaken the  $\text{LiClO}_4$  bond and thus increase the concentration of mobile  $\text{Li}^+$  ions. Figures 6; tables 3; references 14.

**Binary-Nitride Solid Electrolytic Separators for High-Temperature Lithium Storage Batteries**

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Apr 95 Vol 31 No 4, pp 394-402

[Article by N.N. Batalov, O.V. Zheltonozhko, S.N. Zarembo, T.M. Akhmetzyanov, O.V. Volkova, G.V. Zelyutin, V.P. Obrosov, and V.Kh. Tamm, Institute of High-Temperature Electrochemistry at Urals Department, Russian Academy of Sciences, Yekaterinburg; UDC 541.135 (manuscript received 29 Apr 94)]

[FBIS Abstract] The stability of solid electrolytes in contact with lithium is estimated on the basis of a thermodynamic analysis and selection of an adequate separator material for high-temperature lithium storage batteries is proposed. Binary nitrides (lithium nitride and nitride of some II-III-IV group metal) are shown to have the most favorable features: suitable dissociation potential and high corrosion resistance when in contact with lithium cations. This is demonstrated by analysis of polarization processes at the lithium/electrolyte<sub>solid</sub> boundary and at the electrolyte<sub>solid</sub>/electrolyte<sub>melt</sub> boundary. A laboratory model of a  $\text{Li}/\text{Li}_3\text{AlN}_2/\text{LiCl-KCl} + 5 \text{ wt.\% NiCl}_2$  storage battery was built for experimental verification: a vertical BeO cylinder containing  $\text{LiCl-KCl} + \text{NiCl}_2$  melt inside a BeO crucible containing lithium melt, with a  $\text{Li}_3\text{AlN}_2$  plug at the bottom of the cylinder separating the two melts and with a nickel current lead in each melt. The battery was discharged in an argon atmosphere at  $400(+/-)5^\circ\text{C}$ , also while it was being thermally cycled: cooling to room temperature and re-heating to  $400^\circ\text{C}$ . With a 100 mA current flowing during discharge, the battery voltage eventually dropped about 40%. The cause of this deterioration was found to be breakdown of porous nickel in the  $\text{NiCl}_2$  medium. The authors thank G.K. Moiseyev for calculations made pertaining to the thermodynamic aspects of the problem. Figures 7; tables 2; references 43.

**Effect of Size Factor on Transport Properties of Solid Electrolytes with  $\gamma\text{-Li}_3\text{PO}_4$ -Type of Structure**

957A0702J Moscow ELEKTROKHIMIYA in Russian  
Apr 95 Vol 31 No 4, pp 403-405

[Article by Ye.I. Burmakin, G.Sh. Shekhtman, and Ye.R. Aparina, Institute of High-Temperature Electrochemistry at Urals Department, Russian Academy of

Sciences, Yekaterinburg; UDC 541.135.4 (manuscript received 6 Apr 94)]

[FBIS Abstract] An experimental study of the high-conductivity solid electrolytes of the  $\text{Li}_x\text{ZO}_4\text{-Li}_x\text{EO}_4$  system ( $\text{Z} = \text{Si, Ge}$ ;  $x = 2, 3$ ; E- pentavalent or hexavalent element) was made concerning their mechanism of charge transport. Considering that lithium orthogermanate has a higher conductivity than lithium orthosilicate but that the larger  $\text{Ge}^{++++}$  cation is less electronegative than the smaller  $\text{Si}^{++++}$  cation, it would seem that the size factor rather than the covalence factor is responsible for the increase of conductivity as Ge displaces Si. Two systems were selected for verification of this hypothesis:  $0.6\text{Li}_x\text{Si}_{1-x}\text{Ge}_x\text{O}_4$  and  $0.6\text{Li}_x\text{SiO}_{1-x}\text{Ge}_x\text{O}_4\text{-}0.4\text{Li}_x\text{VO}_4$ . As the Ge content (x) was being increased from 0 to 1, all specimens of the first system but only specimens of the second system with  $x > 0.2$  were found to have a  $\gamma\text{-Li}_3\text{PO}_4$  structure. All specimens had been sintered and compacted to a not lower than 90% theoretical density. Measurements were made at temperatures covering the  $250\text{-}750^\circ\text{C}$  range. The temperature dependence of electrical conductivity was found to be typical of such structures with a rigid lattice, phase transition within the  $500\text{-}600^\circ\text{C}$  range being attended solely by relocation of interstitial  $\text{Li}^+$  cations and the trend of this dependence in the high-temperature range corresponding to a consistent decrease of the activation energy. The results indicate that  $\text{Ge}^{++++}$  cations replace  $\text{Si}^{++++}$  cations clusterwise, all the clusters in the first system participating in the transport of charges probably as soon as  $x > 0$  while those in the second system not significantly participating until the Ge content exceeds the  $x=0.6$  level. Figures 2; references 14.

**Principles of Activating Nickel Hydroxide Electrode**

957A0702K Moscow ELEKTROKHIMIYA in Russian  
Apr 95 Vol 31 No 4, pp 406-413

[Article by O.G. Malandin and B.B. Yezhov, Scientific Research Institute of Chemistry at Saratov State University; UDC 541.138 (manuscript received 4 Jul 94)]

[FBIS Abstract] The operation of Ni-Cd and Ni-Fe(Cd) alkaline batteries is analyzed, particularly after activation of their nickel electrode. Although a battery with a  $\gamma\text{-NiOOH}$  electrode discharges at a lower potential and with more intense polarization than does one with a  $\beta\text{-NiOOH}$  electrode, its electrochemical energy conversion capacity may be higher owing to a higher (3.7) attainable oxidization index. While the  $\beta\text{-NiOOH}$  phase and the  $\text{Ni}(\text{OH})_2$  phase are isostructural, their parallel  $[\text{NiO}_2]$  layers are differently ordered and stacked even without interlayer water containing electrolyte cations in neither of them. It therefore is desirable to stimulate either indirect  $\beta\text{-Ni}(\text{OH})_2 \rightarrow \beta\text{-NiOOH} \rightarrow \gamma\text{-NiOOH}$  or direct

$\beta\text{-Ni(OH)}_2 \rightarrow \gamma\text{-NiOOH}$  transition, which can be done by addition of an activator such as  $\beta\text{-Co(OH)}_2$ . Batteries with a ceramic nickel electrode in 1 M KOH electrolyte, some without and some with  $\text{Co(OH)}_2 + 3$  wt.% Co/Ni were not only examined for chemical and phase composition by infrared spectroscopy, Raman-scattering spectroscopy, and x-ray diffractometry but their layer structure was examined by porometry using mercury. The results indicate that especially defectiveness of the  $\gamma\text{-NiOOH}$  phase but also retardation of its formation are the factors which govern the degree of its conversion into  $\beta\text{-Ni(OH)}_2$  during battery discharge. Boosting its defectiveness while the anionic inner structure as well as the surface structure of such a battery is being activated by a Co(II) compound will evidently intensify conversion into  $\beta\text{-Ni(OH)}_2$  and thus raise the battery's discharge capacity. Recovery and retention of the high Ni(II) defectiveness typical of  $\beta\text{-Ni(OH)}_2$  during discharge are, moreover, what ensures excellent performance during a long cyclic operation. Figures 6; tables 5; references 40.

#### Macrokinetics of Oxygen and Hydrogen Cycles in Hermetic Storage Batteries

957A0702L Moscow ELEKTROKHIMIYA in Russian  
Apr 95 Vol 31 No 4, pp 414-419

[Article by Ye.A. Khomskaya, A.L. Lvov, N.F. Burdanova, and N.F. Gorbacheva, Scientific Research Institute of Chemistry at Saratov State University imeni N.G. Chernyshevskiy; UDC 541.138 (manuscript received 11 Apr 94)]

[FBIS Abstract] Both oxygen and hydrogen cycles during charge of new hermetic alkaline and lead storage batteries are analyzed, the purpose being to find a way of accelerating the gas transport. While an alkaline battery is being charged, oxygen accumulates on the positive NiO electrode at an increasing rate. It can be removed either directly by the negative Cd electrode or by a third electrode. Acceleration of the oxygen cycle has been found to be effected by a change of the transport mechanism within the interelectrode space, associated with filtration of the gas which takes place during its diffusion through the separator pores after the latter have been cleared of electrolyte. On a cadmium electrode then appear "gaseous" segments under thin films of the electrolyte solution and these segments become the principal contributors to the oxygen ionization current, owing to the high current density and despite oxygen saturation in the liquid segments. Polarization curves of oxygen ionization at porous Cd electrodes in 5.8 N KOH solution, when fully immersed and when containing absorbed gas, also at porous Pb electrodes in  $\text{H}_2\text{SO}_4$  solution indicate that the oxygen ionization current in-

creases very steeply as the pores become filled with gas. On the basis of measurements has, moreover, been established the dependence of the rate of oxygen recovery at such electrodes on both the surface area of gas absorbing pores and the gas level in them. Calculated current transients attending the initial increasing of the gas penetration depth are compared with oscillograms of the oxygen ionization currents at porous Cd electrodes and indicate a correlation. Hydrogen accumulates on the negative Cd or Pb electrode, from where it can be removed either directly by the NiO cathode or less efficiently and more costly by a noble-metal third electrode acting as catalyst. Measurements made for determining the effect of anodic polarization at a rotating  $\text{PbO}_2$  ring electrode on the rate of hydrogen ionization at that electrode have yielded Tafel curves. Measured was also the hydrogen ionization current at porous  $\text{PbO}_2$  electrodes at various rates of hydrogen intake. A degree of qualitative correlation has been established between pressure transients in the interelectrode space and gas ( $\text{O}_2, \text{H}_2$ ) ionization current transients. An analysis of the results indicates that in order to ensure and regulate forced gas feed so as to maintain a constant pressure in the interelectrode space during charge, it is above all necessary to minimize all clearances during the assembly of the so that they will not exceed the size of their pores. It is also necessary to match the structure of the separator matrix with the structures of the of the porous electrodes so as to facilitate the entry of gas into them. Figures 7; tables 1; references 37.

#### Residual Nitrate-Ion Content and Its Bond Strength in NiO Cermet Electrode

957A0702M Moscow ELEKTROKHIMIYA in Russian  
Apr 95 Vol 31 No 4, pp 420-422

[Article by M.S. Zubov, A.K. Kirakasyan, G.N. Shashkova, and A.A. Bacheva, State Scientific-Industrial Enterprise "Kvant" [Quantum], Moscow; UDC 541.136 (manuscript received 21 Apr 94)]

[FBIS Abstract] The active material in NiO cermet electrodes for alkaline storage batteries is obtained by impregnation the cermet with  $\text{Ni}(\text{NO}_3)_2$  solution and subsequent treatment with caustic potash prior to the molding process. The nickel hydroxide produced by the attendant  $\text{Ni}(\text{NO}_3)_2 + 2\text{KOH} \rightarrow \text{Ni}(\text{OH})_2 + 2\text{KNO}_3$  exchange reaction precipitates in pores of the cermet matrix, where it becomes electrochemically active. Both excess KOH and the niter  $\text{KNO}_3$  are washed off the electrode after molding, but some nitrate ions  $\text{NO}_3^-$  remain. Being a depolarizer, a nitrate ion contributes to corrosive destruction of the nickel base during prolonged battery operation: much more than does a residual  $\text{SO}_4^{2-}$  ion after impregnation with

$\text{NiSO}_4$  solution. Thus arises the problem of inspection for residual nitrate ion during fabrication of these electrodes. Two methods of such an inspection have been proposed: 1. direct potentiometric determination of the nitrate ion in a water extract from the electrode by means of a nitrate-selective electrode ("Kvant" product EIM-11), 2. reduction of the nitrate ion in the electrode to ammonia by Devarda's alloy in an alkaline medium, followed by absorption of the ammonia  $\text{NH}_3$  by an acid and titration of the excess acid or colorimetry with Nessler's reagent. Both methods were tested on electrodes after impregnation and after molding. In a water extract from a  $\text{NiO}$  cermet electrode the  $\text{NO}_3^-$  ion content is one to two orders of magnitude higher than the  $\text{Ni}^{++}$  ion content and two to three times lower than the  $\text{K}^+$  ion content, while the water between N-O layers exists in both "free" and "bound" states. Evidently, therefore, in a water extract from a molded electrode the  $\text{NO}_3^-$  ions are mostly bound to  $\text{K}^+$  ions and the excess  $\text{K}^+$  ions constitute an impurity adsorbed by potassium hydroxide KOH. It is possible, however, that after molding some  $\text{NO}_3^-$  ions will be bound to residual basic nickel salts. Both methods were found to be effective. Tables 3; references 16.

**Charge Voltage Maxima during Operation of Alkaline Storage Batteries with Silver Electrode**  
957A0702N Moscow ELEKTROKHIMIYA in Russian  
Apr 95 Vol 31 No 4, pp 423-430

[Article by I.Ye. Yablokova, Ye.V. Udalova, and G.Z. Kazakevich, State Scientific-Industrial Enterprise "Kvant" [Kvant], Moscow; UDC 541.136 (manuscript received 4 Jul 94)]

[FBIS Abstract] An experimental study of Ag-Zn alkaline storage batteries was made concerning voltage surges during transition from lower-level  $\text{Ag}_2\text{O}$  formation to upper-level  $\text{AgO}$  formation (change of silver from monovalent to bivalent state), such surges also occurring during switching from charge to discharge and from discharge to charge. Tests were performed with smooth foil electrodes 18 mm in diameter and with porous 1.2 mm thick 35x65 mm<sup>2</sup> large sheet electrodes in an open cell containing 10 M KOH solution, also in closed cells. Two  $\text{AgO}$  counterelectrodes were placed one on each side of the Ag electrode and a  $\text{HgO}$  electrode served as reference electrode. Polarization curves were plotted in the galvanostatic mode, a Servogor recording voltmeter with a high input resistance being used for measuring the potentials. Two identical charge-discharge devices were used for instantaneous switching from cathodic polarization with one current to anodic polarization with another current and for instantaneous polarization reversals. The internal resistance of batteries was determined from impedance measurements

made with a 0.1 Hz alternating current and including measurement of the phase angle. The phase composition of Ag electrodes was determined in a DRON-3 x-ray diffractometer using a  $\text{CuK}_\alpha$ -radiation source and a Ni filter. Their structure was examined under an electron microscope with 200-2000x magnification. Voltage surges were found to occur during switching from discharge to charge within the transient period, also during current turn-on or large current rises at the upper level after storage, the duration of the voltage peaks being usually of a longer duration than the "transient" peaks. The maximum potentials increased when the current decreased and when a battery had been precharged to a potential approaching the steady-state lower-level potential. The maximum potential increased as the charge current increased within its low (1-2 mA/cm<sup>2</sup>) range and eventually reached a limit as the current increased further, the magnitude of the surge (difference between maximum and steady-state potentials) thus first increasing and then decreasing. The magnitude of the surges were also found to be higher after longer soaking of the electrode in KOH solution and the maximum surge potential was found to increase after a battery had been charged at a lower temperature. The maximum surge potential decreased slightly during prolonged cycling of a battery, but it was rising faster after battery storage at a higher temperature. Three methods of reducing or even eliminating such voltage surges were successfully tried: a. increasing the number of  $\text{AgO}$  crystallization centers by means of additives, most effective in lowering the maximum Ag electrode potential having been some metal oxides ( $\text{CuO}$ ,  $\text{Bi}_2\text{O}_3$ ,  $\text{PbO}_2$ ) and some tungstanates ( $\text{FeWO}_4$ ,  $\text{NiWO}_4$ ,  $\text{ZnWO}_4$ ); b. diverting some current through a shunt electrode (Cu-Zn) or another Ag-Zn storage cell connected in parallel; c. adding  $\text{PbO}$ , known to decrease the electrode resistance and raise the charge limit prior to liberation of oxygen. In all probability  $\text{PbO}$  also increases the electrical conductivity of the active medium, thus widening the reaction space in the cell and consequently lowering the real current density. Figures 8; tables 1; references 21.

**New Electrochemical System for Nonmetallic Hermetic 1.5 V Storage Battery**

957A0702O Moscow ELEKTROKHIMIYA in Russian  
Apr 95 Vol 31 No 4, pp 431-436

[Article by V.Z. Barsukov, Institute of General and Inorganic Chemistry at Ukrainian National Academy of Sciences, Kiev; I.V. Barsukov and T.I. Motronyuk, Kiev Polytechnic Institute; F. Beck, University of Duisburg (Federal Republic of Germany); UDC 541.136 (manuscript received 21 Apr 94)]

[FBIS Abstract] A new reversible electrochemical system is proposed for nonmetallic hermetic 1.5 V storage batteries with graphite and anthraquinone electrodes,

the problem being to ensure long cycling life without harm to the environment. Experiments were performed with a cathode containing 60 wt.% analytically pure anthraquinone and an anode containing 88 wt.% graphite flakes. Both electrodes had been sintered with an admixture of acetylene black and molded into pellets, anthraquinone without a binder and graphite with a 2 wt.% fluoroplastic emulsion. The current leads were either graphite fibers or, for precision measurements, platinum wires. Each electrode was tested with a Pt counterelectrode and a Ag/AgCl reference electrode. In order for an anthraquinone electrode to function, it is necessary that certain hydrogen ion concentration be maintained so that the main current generating  $AQ + 2H^+ + 2e^- \rightarrow AQH_2$  reaction (AQ- anthraquinone,  $AQH_2$ -anthrahydroquinone) can take place. Another problem is that this reversible reaction may be accompanied by the irreversible disproportioning  $2AQH_2 \rightarrow AQ +$  anthron reaction, anthron being an anthraquinone conversion product not capable of participating in reversible redox reactions. The optimum range of hydrogen ion activity has been found to be  $pH = 0(+/-)0.27$ , the AQ electrode breaking down for increasingly long cycling periods before recovering and finally ceasing to be electrochemically active as the  $pH = 3$  is approached. In search of the best suitable electrolyte, tests were performed not only with conventional  $H_2S$  solution but also with solutions of  $HClO_4$ ,  $HBF_4$ , and other strong acids. The best results were obtained with tetrafluoroborane, an about 80% galvanic utilization factor being attainable with 1-6 M  $HBF_4$  solutions. Adequate performance of the graphite electrode requires, however, a still higher anion concentration. Combining this acid with solutions of tetrafluoroborate salts as additives is helpful in raising the theoretically attainable specific energy content of such a battery beyond 60 (W.h)/kg, but at this time only 18-30 (W.h)/kg seems to be practically realizable. Figures 5; references 10.

**Porous Electrodes and Prototypes of Storage Battery Working with New Electrochemical System Graphite/HBF<sub>4</sub>/Anthraquinone**

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[Article by V.Z. Barsukov, Institute of General and Inorganic Chemistry at Ukrainian National Academy of Sciences, Kiev; I.V. Barsukov and T.I. Motronyuk, Kiev Polytechnic Institute; F. Beck, University of Duisburg (Federal Republic of Germany); UDC 541.136 (manuscript received 21 Apr 94)]

[FBIS Abstract] Several prototypes of graphite/HBF<sub>4</sub> anthraquinone (AQ) storage batteries were built with porous rather than solid electrodes. The basic construction was based on the concept of "plug" electrodes in a polyethylene container: each active electrode material

and the separator molded into a polyethylene plug with a 11 mm inside diameter after the current leads had been inserted first. Two kinds of current leads were used: 1. two thin platinum wires wound into flat spirals, simpler to assemble with a reliable electric contact; 2. two cords of intertwined graphite fiber strands, more difficult to assemble and giving rise to a larger voltage drop but making the battery completely nonmetallic. The contact resistance was not much different, though, at low current density within the 3-5 mA/cm<sup>2</sup> range. Two variants of the basic construction were tested: 1. an "open" one intentionally containing an excess of electrolyte and having an extra polyethylene plug with a capillary duct so as to facilitate independent measurement of AQ electrode and graphite electrode potentials. 2. a hermetic one with each "plug" electrode in a separate container and the two containers welded together. The porous AQ electrode contained 20 g of active material: 50% analytically pure anthraquinone + 50% acetylene black. Three different graphite electrodes contained: 1. 0.310 g of natural graphite flakes + only 2% fluoroplastic emulsion as binder, 2. 0.310 g of natural graphite flakes + 0.043 g of SiC whiskers + 2% fluoroplastic emulsion, 3. 0.310 g of graphite flakes + 0.043 g acetylene black + 2% fluoroplastic emulsion. For the purpose of experimental evaluation there were measured: 1. the cyclic current-voltage characteristics of all three graphite electrodes, in 50 mV/s voltage sweeps from  $E=0$  to potentials covering the 1.6-1.0 V range relative to a Ag/AgCl electrode; 2. the galvanostatic charge-discharge characteristics of the AQ electrode (2.5+ h with 5 mA/cm<sup>2</sup> and 2+ h with 15 mA/cm<sup>2</sup> polarizing currents); 3. the galvanostatic charge-discharge characteristics of the battery prototypes ("open" prototype lasting 4.4+ h with 3 mA/cm<sup>2</sup> and 1.2+ h with 10 mA/cm<sup>2</sup> polarizing currents, hermetic prototype lasting 50-60 min with a 10 mA/cm<sup>2</sup> polarizing current. The results indicate that, following a design optimization, it should be feasible to build such storage batteries with a 20-30 (W.h)/kg specific capacity lasting for at least 100-200 cycles. Figures 7; references 7.

**Characteristics of Zinc-Ferricyanide Redox Storage Batteries**

957A0702Q Moscow ELEKTROKHIMIYA in Russian  
Apr 95 Vol 31 No 4, pp 443-446

[Article by N.D. Koshel, V.G. Zvychaynyy, and N.D. Zaderey, Ukrainian State Chemical Engineering University, Dnepropetrovsk; UDC 541.132.6+ 621.352 (manuscript received 4 Apr 94)]

[FBIS Abstract] The performance characteristics of Zinc-Ferricyanide redox storage batteries with an alkaline electrolyte are overviewed, these batteries hav-

ing a nominal voltage rating of 1.88 V with 64 (A.h)/kg theoretical specific capacity and 117 (W.h)/kg theoretical specific energy. The equation which describes the resultant chemical process in those being developed in the U.S.A. since 1978 is  $Zn + 2K_3[Fe(CN)_6] + KOH = 2K_4[Fe(CN)_6] + Zn(OH)_2$ . Experimental studies were made using not only cells with circulating electrolyte but also half-cells, with 0.5 mm or 1 mm thick carbon-graphite cloth cathode sticking to a 0.27 mm thick cationite membrane in 2.5 M NaH. Local polarization characteristics of such a cathode and charge-discharge characteristics of cells reveal the nature of the electroic processes, particularly important being the anodic process. Inasmuch as the steady-state zinc potential is much more negative relative to a hydrogen electrode in the same medium, there arise several problems: 1. Liberation of hydrogen during the charge cycle lowers the efficiency of this system; 2. Dendritic sediment with a large developed surface in contact with the electrolyte solution corrodes appreciably fast with attendant release of hydrogen; 3. Emergence of hydrogen precludes the possibility of hermetically sealing the circulation system so that other means are needed for protection of the alkaline electrolyte against leakage loss and prevention of its coming in contact with the ambient environment; 4. Formation and entrapment of hydrogen bubbles within the dendritic sediment increases appreciably the resistance of the electrode and thus also the charge voltage. All this is reflected in the current-voltage and current-power characteristics measured with electrolyte flowing through the anodic loop at rates 0.012-0.023+ cm<sup>3</sup>/(s.cm) and through the cathodic loop at rates of 0.25-0.34 cm<sup>3</sup>/(s.cm). On the basis of these data are estimated the potential capabilities of a Zn-Fe(CN)<sub>6</sub> redox system: current density 0.09 A/cm<sup>2</sup>, power density 0.092 W/cm<sup>2</sup> (3.6 mm thick cells), specific power about 90 W/kg (mean mass of working medium 3 g/cm<sup>3</sup>). Figures 3; references 15.

#### Possibility of Gyroscopic Stabilization of Rotation of System of Solid Bodies

957A0957A Moscow PRIKLADNAYA MATEMATIKA I MEKHANIKA in Russian  
Vol 59 No 3, May-Jun 95 pp 385-390

[Article by A. Davyskib and V.A. Samsonov, Sydney/ AUSTRALIA and Moscow; UDC 531.36:62-50 (manuscript received 1 Apr 94)]

[FBIS Abstract] The possibility of gyroscopic stabilization of a spacecraft's permanent rotation of a spacecraft is analyzed, such a generally multidimensional multi-parametric object being reduced to the simplest model: an axisymmetric main body in the form of cylinder (radius R) rotating about its axis of symmetry OZ while

fixed at some point on that axis and coupled through two frictionless cylindrical hinges to two identical rods on the circumference around that fixed point. This system of three bodies has five degrees of freedom and its position in a stationary three-dimensional Cartesian system OXYZ is described by five coordinates: two Krylov angles defining the orientation of the OZ cylinder axis in some moving Cartesian system OZ<sub>1</sub>x<sub>1</sub>y<sub>1</sub> which rotates about the OZ axis of the cylinder and two angles defining the positions of the two rods relative to the cylinder. There are assumed to be no external forces acting. On this basis is first established the condition for stability relative to the equilibrium of a rod and is examined the behavior of the positioning forces. In accordance with Kelvin-Chetayev theorem, gyroscopic stability is shown to be possible in only two out of the ten different regions of the l,b-plane: l= L/R, b= (I<sub>1</sub>/I<sub>3</sub>)- 1; I<sub>1</sub>,I<sub>3</sub>-equatorial and axial moments of inertia. Projecting its kinetic moment G onto the two O<sub>1</sub>x<sub>1</sub>,O<sub>1</sub>y<sub>1</sub> axes and using the particular integrals G<sub>1</sub>(t),G<sub>2</sub>= 0 can, generally, change the nature of the stability problem: from stability throughout the entire phase space to stability on some manifold. In this special case, however, both problems are evidently equivalent. Figures 3; tables 1; references 5.

#### Design of Optimum Nose Contour for Immersion in Supersonic Stream

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Vol 59 No 3, May-Jun 95 pp 419-434

[Article by A.N. Krayko and D.Ye. Pudovilov, Moscow; UDC 533.6.011 (manuscript received 5 Oct 94)]

[FBIS Abstract] The problem of profiling the nose contour of a flat body so as to ensure minimum wave drag along the entire length of the body in a uniform oncoming supersonic stream of a nonviscous and thermally nonconducting ideal gas. The problem is formulated in Cartesian coordinates with one of the axes running in the direction of the stream. Given are the Mach number or velocity V<sub>∞</sub> of the oncoming stream and the relative thickness  $\tau > 1$  of the body. The problem is treated as a variational one for the system of Euler equations, its linear part having already been solved by the method of variation in characteristic  $c^+$ -planes. This method is now applied to the part of the nose contour along which an attached shock wave will flow. The solution to the problem depends largely on the reflection coefficient  $\lambda(V_\infty, \tau) = \delta p^+ / \delta p^*$  ( $\delta p^+$  denoting pressure perturbations at jumps along the  $c^+$  characteristics. Considering that this part of the contour is quasi-straight, a wedge formed by a straight generatrix is shown to be the optimum profile when

$\lambda = 0$ . The "principal correction", breaks of a straight generatrix, is then obtained when  $\lambda > 0$  and when  $\lambda < 0$ . The theoretical solution to this problem is supplemented with a numerical analysis. The authors thank V.A. Vostretsova for assistance. Figures 4; tables 1; references 15.

**Motion of Body Through Large-Scale Inhomogeneity in Stratified Atmosphere**  
*957A0957C Moscow PRIKLADNAYA MATEMATIKA I MEKHANIKA in Russian*  
*Vol 59 No 3, May-Jun 95 pp 435-441*

[Article by V.U. Nabiiev, S.V. Utyuzhnikov, and N.K. Yamaleev, Moscow; UDC 533.6 (manuscript received 25 Jun 93)]

[FBIS Abstract] Motion of a solid body entering the atmosphere through a cloud is considered, the trajectory of such a body depending on the aerodynamic coefficients and the latter being usually calculated for solving the problem of streamline flow. This requires a simultaneous solution of the differential equations of motion for the body and the differential equations of gas flow past the body. In order to obtain an accurate solution, it is necessary that these equations be integrated over sufficiently small discrete successive time intervals. For long bodies of revolution moving freely at a supersonic speed and usually at small angles of attack a better method is the method of a small parameter. With the angle of attack  $\alpha$  selected as the small parameter, the dependence of any other parameter on it can then be expressed in an analytical form. The method is applied to bodies entering the atmosphere of Earth, a stratified medium, after a cloud of hot gas has formed in it with a temperature distribution  $T(z,r) = T_a(z) + (T_{max}(z)\exp[-(l/R_T)^2])$  and evolved into a toroidal vortex while buoying up:  $T(a)$ - temperature of quiescent atmosphere at altitude  $z$ ,  $r$ - radial coordinate,  $T_{max} = 10,000K$  (at altitude  $H = 20$  km),  $l$ - distance from center of spherical space,  $R_T = 4$  km. The flow of gas past the body is described by complete shock layer equations. The three-dimensional solution to the equations of motion is expanded into an asymptotic power series in that small parameter (angle of attack). The coefficients of this series are expanded into a formal Fourier series in the meridional angle  $\psi$ , only the two terms  $\Phi_0 + \alpha\Phi_1 \cos\psi$  being retained and calculated by the method of global iterations. There are then calculated the drag coefficient, the lift coefficient, and the pitching moment - all not any more as functions of the angle of attack. The trajectory and the spatial orientation of the body are determined from the matrix system of equations  $d[\xi]/dt = [F]([\xi], [Q], [A])$  (G. Martin; "Entry into the Atmosphere", Izd. Mir, Moscow 1969). The problem has been solved numerically on an IBM

386/387 computer by the method of successive approximation using the Runge-Kutta scheme of third-order precision with respect to intervals of the flight time. It has been solved for two cones entering a cloud with a velocity of 2000 m/s at an instant 15 s after the latter had been lifted by Archimedes' force to an altitude of 22 km: each cone being 2 in long and having a 30° vertex angle, a spherically rounded tip (radius 0.1 m), and a mass of 1 ton. The cone with a 50 cm long distance from its center of mass to its vertex has a large stability margin and during its passage through the cloud the angle of attack fluctuates slightly, the presence of a heat source along its path evidently having almost no effect on its trajectory. The cone with a 140 cm long distance from its center of mass to its vertex is less stable and the angle of attack fluctuates widely, the presence of a heat evidently having an appreciable effect on its trajectory. The authors thank G.A. Tirshty for attentiveness and discussion. Figures 6; tables 1; references 5.

**Measurement of Energy Spectrum of Neutrons Generated in "Thick" Aluminum Target by Bombardment with 70 MeV Protons**

*957A0959A Moscow PRIBORY I TEKHNIKA EKSPERIMENTA in Russian*  
*No 3, May-Jun 95 pp 28-33*

[Article by V.D. Sevastyanov, G.B. Tarnovskiy, G.A. Borisov, L.S. Saltykov, Ye.P. Kadkin, and V.I. Lyagushin, All-Russian Scientific Research Institute of Engineering Physics and Radio Engineering Measurements, Moscow; UDC 539.125.52 (manuscript received 14 Apr 94, after completion 9 Nov 94)]

[FBIS Abstract] In a special experiment was measured the energy spectrum of neutrons generated by bombardment of an aluminum target with a proton beam. A beam of 70 MeV protons, extracted from the U-240 isochronous cyclotron in the Institute of Nuclear Research at the Ukrainian Academy of Sciences, was passed through an ion pipe into the interaction chamber. The target inside this chamber was a 22 mm thick plate. Such a thickness, larger than the 20 mm long mean free path for 70 MeV protons in aluminum, ensured that hardly any scattered protons would fall into the neutron detectors. Measurements were made by the neutron-activation method with a set of neutron detectors containing fissionable  $^{235}\text{U}$ ,  $^{238}\text{U}$ ,  $^{237}\text{Np}$ ,  $^{209}\text{Bi}$  nuclides so as to cover the 0.5 eV - 55 MeV range. These detectors were placed inside the chamber behind the target, in a plane perpendicular to the axis of the primary proton beam at a 20° angle to it. Fast neutrons were monitored by a sensor cell containing a suspension of ZnS powder in a hydrogenous medium. The incident proton beam was monitored by means of a 30  $\mu\text{m}$  thin

copper facing it upon its entry into the chamber and a radiometer recording the protons scattered by this foil. The radiometer was placed inside the chamber off the axis of the incident proton beam at a 20° angle to it and had been calibrated against a Faraday cylinder prior to insertion of the target. Behind the foil and before the target was placed an annular collimator absorbing the protons scattered inside the ion pipe and then by the foil. The thickness of the neutron detectors varied over the 0.5-2 mg/cm<sup>2</sup> range, based on the maximum sensitivity-to-cost ratio of each. Their sensitivity was varied by means of various shields (1 mm thick Cd layer, 0.4 g/cm<sup>2</sup> and 1.0 g/cm<sup>2</sup> <sup>10</sup>B layers). A neutron-activation <sup>127</sup>I detector was added for recording neutrons produced by the <sup>127</sup>I(n,2n)<sup>126</sup> reaction, the energy threshold for this reaction being 10.8 MeV. Measurements were also made by the time-of-flight method. There was, furthermore, directly measured the number of nuclear reactions taking place in each detector during its bombardment by the neutrons coming from the aluminum target. This number has been normalized into the number per one primary proton and one nucleus of the detector nuclide-target. It had been necessary to match the measured numbers of reaction with the calculated ones by different methods above and below the 18 MeV level. Extrapolation of the spectrum below the 3 MeV level was done by sampling available energy spectra of neutrons similarly generated in target materials having mass numbers, such as in an iron target by bombardment with 52 MeV protons, along with one of neutrons generated in an aluminum target by bombardment with 200 MeV protons. Figures 2; tables 1; references 8.

#### Radiophysical Apparatus for Study of Internal Defects in Radiotransparent Specimen

957A0959B Moscow PRIBORY I TEKHNIKA  
EKSPERIMENTA in Russian  
No 3, May-Jun 95 pp 102-106

[Article by A.A. Vertiy, I.Ya. Gudym I.V. Ivanchenko, Yu.V. Karelina, and N.A. Popenko, Institute of Radiophysics and Electronics at Ukrainian Academy of Sciences; UDC 621.373.826 (manuscript received 11 Apr 94, after completion 27 Jul 94)]

[FBIS Abstract] An apparatus has been designed and built for nondestructive detection and location of internal flaws in radiotransparent objects made of ceramic, dielectric, and similar materials. It measures amplitude and phase distributions of the electromagnetic field passing through such an object and thus supplies the necessary information. The radiophysical part of the apparatus consists of a microwave oscillator followed by a directional coupler, an attenuator, and a transmitter antenna before the specimen, with a receiver antenna

behind it. The specimen is attached to the frame of an XYZ-scanner and moved by it in all directions between the two antennas. The measuring system is interfaced with an IBM PC/AT 286 computer. It begins with a directional coupler connected to the receiver antenna, from where one path leads through a ferrite diode, a semiconductor modulator, and a diode to a tuned amplifier which feeds signals through the interface to the computer. This coupler is also followed by a second one, from where one path leads directly to a decoupler which receives signals from a phase modulator and another path leads through a diode to a tuned amplifier which feeds signals to a phase detector. The directional coupler between the oscillator and the attenuator is also connected to a decoupler. This one and a sound generator feed signals to the phase modulator, which then feeds signals to the phase detector. The phase detector and the scanner both feed signals through the interface to the computer. The main technical characteristics of the apparatus are: minimum cross-section of probing microwave beam 0.5x0.5 mm<sup>2</sup>, maximum scanning range 200x200 mm<sup>2</sup> surface area, error of setting the coordinates of a target spot on the specimen surface (+/-)0.005 mm. The electromechanical part of the apparatus consists of five ShD-0.4 stepper motors and 10 terminal switches, four for programmed commutation of the motor windings and five for chopping the power supply. The apparatus is suitable for inspection of materials having a dielectric permittivity  $1 < \epsilon < 16$  and a loss tangent  $\tan \delta < 0.01$  range. The measuring system can operate in three modes: 1. and 2. recording the amplitude distribution or the phase distribution of the electromagnetic field, for detection of internal flaws; 3. simultaneously recording both distributions, for structural examination of the resonance vibration modes in open cavities. The control program for this apparatus has been written in the TURBO-PASCAL version 7.0 language, fragments of it in the TURBO-ASSEMBLER version 2.0 language, for a version 6.0 disk operating system. Figures 4; references 3.

#### Spectrometer of Coherent Anti-Stokes Scattering for Isotope Analysis of Hydrogen in Thermonuclear Facilities

957A0959C Moscow PRIBORY I TEKHNIKA  
EKSPERIMENTA in Russian  
No 3, May-Jun 95 pp 172-179

[Article by A.A. Ivanov, G.A. Polyakov, and V.B. Voronin, Institute of Innovation and Thermonuclear Research, Troitsk (Moscow Oblast) (manuscript received 30 Mar 94, after completion 10 Nov 94)]

[FBIS Abstract] A spectrometer has been developed for quantitative control of the fuel mixture in fusion reac-

tors by monitoring the content of hydrogen isotopes in it. The spectrometer operates by the method of coherent anti-Stokes scattering of light, a parametric four-photon process in a medium having a cubically nonlinear susceptibility where a coherent directional radiation with an anti-Stokes frequency  $\omega_{as} = 2\omega_o - \omega_s$  is generated by interaction of Stokes radiation (frequency  $\omega_s$ ) and reference radiation (frequency  $\omega_o$ ). In the absence of one-photon and two-photon absorption the susceptibility of that medium is the sum of two components representing respectively the sought molecular resonance  $\omega_o - \Omega$  (frequency of Raman-active transition) and the electronic as well as other far-off-resonance transitions. The advantages of this method are: 1. high spectral resolution, limited only by widths of the two radiation bands; 2. strong recorded signal; 3. small divergence angle comparable with that of lasers beams, owing to phase coherence; 4. high spatial resolution, owing to a small two-wave interaction space. The spectrometer for quantitative analysis of hydrogen isotopes uses stimulated Raman scattering in high-pressure  $^1\text{H}$  or  $^2\text{H}$  as the way to generate the pumping Stokes radiation. The light source for this spectrometer is a pulsed YAG:Nd laser (Quantel International YG581-10 model) with Q-switching, followed by a LiNbO<sub>3</sub> frequency doubler (second-harmonic generator) crystal which emits the 532 nm reference radiation in pulses of up to 350 mJ energy and of 10 ns duration. The spectrometer itself includes an SRS (stimulated Raman scattering) cell filled with hydrogen isotope gas under a pressure of 1.5-25 atm, an interaction cell containing the hydrogen-deuterium mixture and having three inserted temperature sensors, four biconvex focusing lenses, four filters, three beam splitter plates with 10% reflectance, and two rotating mirrors, three calorimeters, a polychromator and a 5-channel optical analyzer of the coherent anti-Stokes scattering signal (Princeton Applied Research "IG&G" OMA-2 model: resolution 2 nm, sensitivity 20 photons/reading) placed behind the polychromator with a 1 nm/channel dispersion. One calorimeter measures the energy of the 1.06  $\mu\text{m}$  laser emission pulses, two calorimeters measure the energy of reference radiation pulses and Stokes radiation pulses respectively. During measurements the  $\text{H}_2\text{-D}_2$  mixture in the interaction cell is under its full pressure of 50 torr. Prior to each successive set of measurements the gas supply line and the interaction cell, both made of stainless steel, are heated to 250°C by a flexible ribbon heater and then vacuumized by a fore pump acting through a set of nitrogen traps so as to minimize the effects of adsorption and desorption. The output signals from the two reference radiation and Stokes radiation calorimeters are sent to a storage oscilloscope. This instrument and the optical analyzer are coupled to a data acquisition and processing

IBM PCVAT programmable control computer. Figures 4; references 17.

#### Controllable Isotopic Sources of Fission Fragments Based on Nuclear Physics Facilities

957A0969A *Moscow PRIBORY I TEKHNIKA EKSPERIMENTA* in Russian  
No 3, May-Jun 95 pp 25-27

[Article by V.D. Sevastyanov and G.N. Maslov, All-Russian Scientific Research Institute of Engineering Physical and Radiotechnical Measurements, Mendeleyevo (Moscow Oblast); UDC 539.173.8.-3:001.8 (manuscript received 10 Feb 94)]

[FBIS Translated Text]

#### Summary

On the basis of a neutron generator and a BIR-2 nuclear reactor have been produced isotopic sources of fission fragments suitable for calibration of fission fragment detectors. These sources consist of radiators with fissionable  $^{235}\text{U}$  nuclide located either in the field of thermal neutrons in the moderator assembly which slows down 14 MeV neutrons, or in the isotropic field of fast neutrons in the middle of the reactor core. The fragmentation activity of these sources is tracked either on the basis of readings given by a counter of  $\alpha$ -particles attending the emission of 14 MeV neutrons (from the neutron generator) or readings given by a monitor of the energy release in the BIR-2 reactor core, measurable in the fission chamber.

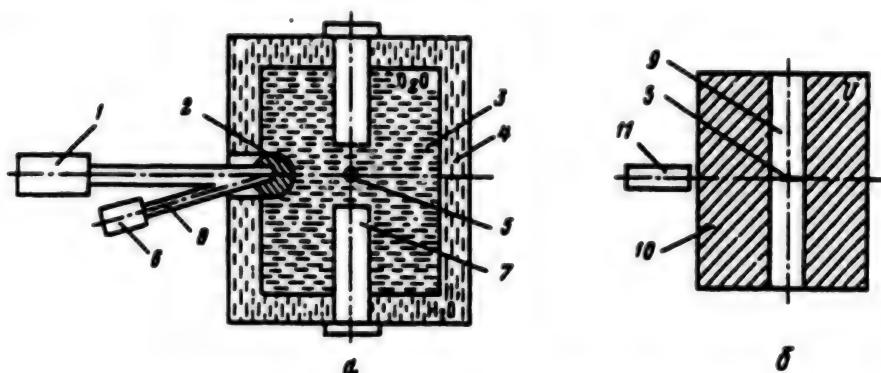
Fission fragment sources are used for calibration of fission fragment detectors, in production of nuclear filters, and for other purposes. [1-5]. Fission fragment sources based on spontaneous fission of  $^{252}\text{Cf}$  and  $^{244}\text{Cm}$  nuclides are being produced in Russia since 1977. However, the fragmentation activity of such sources is low (100 Bq) and their guaranteed service life is short (up to 1 yr).

This article reports the results of work done on constructing two isotopic sources of fission fragments: IOD-1 based on a neutron generator and IOD-2 based on a BIR-2 nuclear reactor having an open metallic core. [Istochnik Oskolkov Deleniya = Source of Fission Fragments].

The IOD-1 (Figure a) contains a generator of 14 MeV neutrons (1) a converter (2) made of lead, heavy water (3) serving as moderator, water bidistillate (4) serving as reflector, a fission fragments radiator (5) containing the  $^{235}\text{U}$  nuclide, a counter (6) of  $\alpha$ -particles attending the emission of 14 MeV neutrons, and a graphite plug (7). The nuclear  $\text{T}(\text{d},\text{n})^4\text{He}$  reaction in the target slug (8) of the generator (1) produces 14 MeV neutrons. In the spherical converter with a 130 mm outside diameter these neutrons are, by way of  $(\text{n},\text{n}')$  and  $(2\text{n},\text{n}')$  reactions, changed into 0.8-2 MeV neutrons.<sup>6</sup> In the

moderator (3), which forms a 524 mm high cylinder with a 3500 mm outside diameter, these  $0.8 \div 2$  MeV neutrons are slowed down to become thermal ones. Escape of thermal neutrons is minimized by placement of the moderator inside a reflector (4), a 810 mm high hollow cylinder with a 900 mm outside diameter. Fission fragments are produced by the thermal neutrons

in the  $^{235}\text{U}(\text{n},\text{f})$  reaction in the radiator (5). There is a one-to-one relation between the fragmentation activity (f-activity) of the radiator and the rate of pulse count by the counter of ga-particles attending the emission of 14 MeV neutrons.<sup>7</sup> The fission fragments emanate from the layer of fissionable radiator (5) material isotropically over a solid angle of  $4\pi$  sr.<sup>8</sup>



Схемы источников осколков деления: ИОД-1 (а). ИОД-2 (б)

Configurations of Fission Fragment Sources: IOD-1 (a), IOD-2 (b)

The configuration of source IOD-2 is shown in Figure b. A radiator (5) containing a  $^{235}\text{U}$  layer is situated at the geometric center in the central channel (9) of the BIR-2 reactor core (10). The inside diameter of that central channel is 45 mm. When the reactor is operating there are fast neutrons generated in its core which then, by way of the  $^{235}\text{U}(\text{n},\text{f})$  reaction, produce fission fragments in the fissionable radiator (5) material. The fission fragments emanate from the radiator isotropically over a solid angle of  $4\pi$  sr, duplicating the field pattern of fast neutrons at the center of the core. There is a one-to-one relation between the number of fission events in the radiator and the readings given by the monitor (11) of energy release in the reactor core (fission chamber).<sup>10</sup>

The radiators of fission fragments in both fission fragment sources are layers of fissionable materials with the  $^{235}\text{U}$  nuclide deposited on aluminum substrates in the form of disks. These layers are "thin", their thickness being smaller than the mean length of path of fission fragments in the fissionable material.

The fission fragment radiators in both IOD- and IOD-2 are layers of fissionable material with  $^{235}\text{U}$  nuclide deposited on aluminum substrates in the form of disks. Such a layer is "thin", its thickness being small in comparison with the length of path of fission fragments in the fissionable material. The thickness of the  $^{235}\text{U}$  layer in these radiators is 1 mg/cm<sup>2</sup>. The layers of fissionable material were tested by certain methods<sup>11</sup> for certification as to the number of  $^{235}\text{U}$  nuclei, with an error not larger than 1%.

Measurements of the f-activity in the two sources were made with the aid of small fission cameras and muscovite mica track recorders. The methods of measurement with fission fragment detectors have already been discussed.<sup>1,10,12</sup> Some characteristics of the two sources are given in the Table.

The error of measurement of the quantities listed in the Table does not exceed 2% ( $P=0.95$ ).

IOD type	Mass of $^{235}\text{U}$ in radiator, mg	f-activity during emanation of 14 MeV neutrons at a rate of $1.0 \times 10^9$ n/s from target, Bq	Number of fission events during release of 1 MJ energy in BIR-2 reactor core
IOD-1	1	97.5	-
IOD-2	1	-	$5.13 \times 10^8$

It follows from the data in the Table that:

1. By varying the neutron yield from the target slug of the neutron generator, one can regulate the f-activity in the IOD-1 over the  $1 \div 1000$  Bq range;
2. By changing the energy release in the reactor core by  $6 \div 7$  orders of magnitude, one can increase the number of fission events in the radiator over the  $10^3 \div 10^{10}$  range;
3. The two IOD-1 and IOD-2 complement each other on the range of f-activity reproduction.

It should be noted that emission of fission fragments in each IOD is accompanied by multiconstituent emission (neutrons,  $\gamma$ -ray, characteristic x-rays, and others). This property of these sources makes them unique with respect to radiative action on objects under study.

Further improvement of these sources entails:

1. formation of a field of fission fragments in a large volume, for the study of radiative action on large objects, which requires the use of a reactor with a large cavity in its core (about 300 mm)<sup>13</sup> accommodating a radiator with  $^{235}\text{U}$  in the form of a thin-walled hollow cylinder inside which an isotropic field of fission fragment will form;
2. widening the range of f-activity generation by several orders of magnitude, namely using a more power reactor in the IOD.<sup>14</sup>

The upshot of this work is construction of the first Russian devices based on nuclear physics facilities for measuring the fragmentation activity and the number of fission events.

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#### High-Temperature Interaction of U and Zr with Water Vapor

957A0985 Lviv FIZIKO-KHIMICHESKAYA MEKHANIKA MATERIALOV in Russian  
No 3, May-Jun 95 pp 36-43

[Article by V. N. Polyakov, I. P. Gnyp, K. N. Gedkov, G. S. Bulatov, B. M. Kazakov; G. V. Karpenko Physico-mechanical Institute, National Academy of Sciences of Ukraine, Lviv; UDC 537.311.62 (manuscript received 9 Feb 94)]

[FBIS Abstract] When uranium and zirconium oxidize, hydrogen, an explosive gas, is released. This paper examines the high-temperature interaction of uranium and zirconium with water vapor. At temperatures up to 700 K, the kinetics of uranium oxide are governed by a linear law, and above 900 K by a parabolic law. Oxidation of zirconium and an alloy of zirconium (Zr + 1 percent Nb) by water vapor is subject to a cubic law at 900-1200 K and a parabolic law at 1300-1500 K. The electrochemical properties and structural defects of the oxide films that are formed are

used to discuss mechanisms of uranium and zirconium oxidation by water vapor. Uranium oxidation is due to diffusion of OH-anions from the node into the internode space. In zirconium, oxidation is due to anion vacancies. The initial stage of zirconium oxidation is characterized by diffusion of cations to the gas-solid interface. Comparative analysis of the kinetics of the release of hydrogen in high-temperature oxidation of

uranium and zirconium by water vapor shows that the average rate of hydrogen release (as well as other end products) is approximately twice as high in uranium, and is dependent on temperature, the length of contact, pressure, the state of the uranium surface and the presence of admixtures in the vapor and uranium. Figure 1; tables 4; references 6 (Russian).

**Organophosphorus Chemicals Suspected in  
Banker's Murder**  
**957A1088**

[FBIS Media Note] Russian media reports indicate that organophosphorus chemicals may have been used to poison prominent banker Ivan Kivelidi, allegedly murdered on 4 August. Circumstances suggest that the murderer was able to gain access to the victim's office despite his large, well trained security staff (KOMSOMOLSKAYA PRAVDA 11 Aug 95). The Gangsterism and Homicide Department of the Moscow Prosecutor's Office is conducting an intensive criminal investigation (KOMSOMOLSKAYA PRAVDA 11 Aug 95), with results to be released in several weeks (IZVESTIYA 11 Aug 95).

IZVESTIYA (11 Aug 95) and KOMSOMOLSKAYA PRAVDA (11 Aug 95) offer different theories of how organophosphorus chemicals might have been used. According to KOMSOMOLSKAYA PRAVDA, organophosphorus vapors could have been used as a back-up to heavy metal poisoning of the banker's food. In contrast, IZVESTIYA contended that the banker had been killed by organophosphorus powders alone. Both newspapers allude briefly to the fact that this class of chemicals includes military agents, as well as common pesticides.

Kivelidi, chairman of the Russian Business Round-table, was the tenth member of this organization to be murdered during the past year (INFORMATSION-NOYE AGENTSTVO EKHO MOSKVVY 8 Aug 95). Despite this pattern, Kivelidi's symptoms were initially attributed to chronic health problems. Authorities only began to suspect poisoning when Kivelidi's healthy 35-year-old secretary, Zara Ismailova, developed the same symptoms exactly 24 hours after her employer and quickly died. Subsequently two police officers were diagnosed with poisoning after examining Kivelidi's office.

Preliminary medical reports attributed the murders to heavy metals, probably cadmium (IZVESTIYA 5 Aug 95, Itar-TASS 8 Aug 95, ROSSIYSKAYA GAZETA 17 Aug 95) — poisons that, according to Alexey Kovalenko, head of the Laboratory for Analytical Methods, can be administered only via food (KOMSOMOLSKAYA PRAVDA 11 Aug 95). The Sklifosovskiy Institute Poison Center reported that both the banker and his assistant had been poisoned by identical heavy metal salts, probably simultaneously (IZVESTIYA 5 Aug 95). The center reportedly thought that differences in medical treatment, age and health accounted for the secretary's more rapid demise (IZVESTIYA 5 Aug 95).

IZVESTIYA reporter Kirill Svetitskiy, however, theorized that organophosphorus chemicals alone had been used. He reasoned that since heavy metals are readily detected by spectrography, the unusually protracted forensic investigation probably meant that no heavy metals had been found (IZVESTIYA 15 Aug 95). According to Svetitskiy, only organophosphorus compounds could explain the case. Unnamed medical "experts" reportedly told him that the victims' symptoms "unambiguously" describe "rapid brain edema caused by an acute hypertensive crisis" [a diagnosis echoed by KOMSOMOLSKAYA PRAVDA (11 Aug 95)]. They reportedly said that only other substances capable of causing these symptoms were synthetic analogues of adrenaline or noradrenaline, which neither the victims nor the poisoned investigators had taken. Furthermore, Svetitskiy noted properties of organophosphorus chemicals that would make them attractive murder agents: they are relatively stable in powdered form, do not break down at room temperature, and lack a sharp smell. Moreover, he wrote, organophosphorus chemicals cannot be detected in the body two days after exposure; low cholinesterase levels are the only indication of poisoning.

The IZVESTIYA reporter speculated that the murderer knew that Kivelidi, who was on a medically restricted diet, always lunched at his desk, eating sandwiches prepared by his secretary. Svetitsky further theorized that the murderer sprinkled organophosphorus powder on surfaces that the banker was likely to touch in the hopes that he would ingest some of the poison. Svetitskiy speculated that Ismailova was poisoned because she tidied her boss' office, then went to eat. In his analysis, only slight chemical traces remained to poison police on 3 August, while investigators who tried to find organophosphorus chemicals on 11 August found nothing. The credibility of Svetitskiy's theory is undercut by previous articles in which he had reported that Kivelidi never ate in his office (IZVESTIYA 5 Aug, 9 Aug 95).

**Fatal Four O'Clock Slump**

Svetitskiy's theory is based in part on a detailed account of Kivelidi and Ismailova's illnesses provided by Rosbiznesbank vice president Sergey Bachurin (IZVESTIYA 11 Aug 95). Bachurin's information is summarized below and compared to other press reports.

- On 1 August, at 4 pm, two hours after arriving at the office, Kivelidi collapsed and fainted, swallowing his tongue. Colleagues were unable to pry his mouth open to help him. His condition was so severe that "it took an hour and a half to prepare him for transport" to the Central Clinical Hospital. He began to vomit white mucus. The physi-

cians' initial hypotheses were stroke, cerebrovascular thrombosis, or extensive infarct.

- On 2 August, also at 4 pm, Kivelidi's secretary, Zara Ismailova, suddenly began to feel sick and fainted. She was taken by ambulance to the First Municipal hospital where she was treated for epilepsy in the neurology ward. She died at 6 am on 3 August. The physicians diagnosed acute vascular insufficiency as the cause of death, although Ismailova had no history of cardiovascular or other chronic disease.
- On 3 August, a large group of police investigated and sealed Kivelidi's Rosbiznesbank office. Two members of this team developed eye pain similar to the victims' (again at 4 pm according to KOMSOMOLSKAYA PRAVDA 11 Aug 95), and as a result the police evacuated the bank staff from the building. KOMSOMOLSKAYA PRAVDA (11 Aug 95) reported that the Gelmolets Institute for Ophthalmic Diseases concluded that the investigators had been poisoned with an unknown agent.
- On 4 August, the police team returned to the bank together with an expert chemist. Kivelidi died the same day without emerging from a "deep coma."

Meanwhile bank vice president Bachurin had Ismailova's colleagues document everything they could recall of the day she fell ill. They reported that the secretary felt well before lunch and spent some time tidying Kivelidi's office. By 3 pm, her eyes and skin had turned red. At 4:30 she complained of eye pain and headache and began to lose consciousness. Her face and torso became cherry-red. She partially regained consciousness, breathing rapidly. Her eyes teared and protruded somewhat from the sockets with contracted pupils. She said, "I am dying. It's exactly the same thing that happened to Ivan Kharlampiyevich [Kivelidi]." Ismailova said her vision was blurred and she could see nothing. The secretary became disoriented, trying to tear her clothing off and scratching her skin "as if she wanted to remove it." Ismailova alternated between agitation and calm. Her hands twitched convulsively, her face became sweaty and brown, and her tongue enlarged, turned purple and began to slip down her throat. At 17:35, she vomited white mucus, then bile. She was then transported to the hospital where she lapsed into a coma and died.

KOMSOMOLSKAYA PRAVDA gave a similar, though less detailed, account of both victims' conditions (11 Aug 95), writing that Ismailova had "symptoms of acute intoxication: eye pain, convulsions,

vomiting of dark mucus, unconsciousness." The same report noted that Kivelidi had kidney and liver failure, a high temperature and brain edema. SEGODNYA (5 Aug 95) wrote that Ismailova's autopsy detected pulmonary and cerebral edema (symptoms that reportedly indicate rapid death by asphyxiation) and revealed no specific injuries to her internal organs beyond slight bleeding near the pancreas. ROSSIYSKAYA GAZETA (17 Aug 95) reported that Rosbiznesbank representatives suspected radiation poisoning because "many coworkers" had developed the same symptoms as Kivelidi after spending time in his office and "those who used his telephone had to be hospitalized."

#### Official Report Awaited

According to a confidential source within the prosecutor's office (IZVESTIYA 11 Aug 95), forensic and investigation results were to take at least several weeks. Among studies being performed were histological examination of Ismailova's internal organs, analysis of scrapings from objects in Kivelidi's office, and examination of food products taken from his office refrigerator.

#### Summaries of Papers Delivered at Annual Conference on the "Gene and Cell Engineering" Part of the Russian State Scientific and Technological Program: "Latest Bioengineering Methods"

957A0264A Moscow MOLEKULYARNAYA GENETIKA, MIKROBIOLOGIYA I VIRUSOLOGIYA in Russian No 5, Sep-Oct 94 pp 32-41

[Article!]

[FBIS Translated Text]

#### Project 3. Recombinant Bacteria

#### Development of Modern Methods for Detection and Prevention of Brucellosis Using Gene Engineering Methods

Yu. V. Kulakov, D. F. Selyutina, M. M. Zheludkov, Ye. A. Dranovskaya, and A. G. Skavronskaya, RAMS Research Institute of Epidemiology and Microbiology imeni N. F. Gamaleya, Moscow

The brucella genetics team previously created a library of genes of highly virulent strains of *B. melitensis* 565 in *E. coli* cells. Clones were selected that produce specific brucellosis protein antigens with mol. wt. 38 and 18 kD [dalton].

The purpose of these studies was to develop modern techniques and agents for diagnosing and preventing

brucellosis on the basis of products of cloning genetic determinants of brucella in *E. coli*. Preparative isolation of brucella antigens from producer strains, purification and study of their physicochemical and immunobiological properties were carried out in 1993.

It was established that the optimum method of isolating proteins from brucella external membrane proteins from producer strains is cryogenic osmotic shock which yields not only more pure antigens, but also more active ones (RIA [radioimmune assay] titer 1:16).

It was shown that antigens with mol. wt. 38 and 18 kD are notable for thermostability, partial hydrolysis in 0.1 N HCl and total hydrolysis in 0.1 N NaOH, they are not destroyed by pronase, proteinase K, phospholipase and 2-mercaptoethanol. After papain treatment the 18 kD antigen loses its capacity to react with homologous antibodies, whereas 38 kD antigen shows change in electrophoretic mobility and migrates to the anode zone in gel after trypsin treatment.

We explored the possibility of using antigens of brucella synthesized in *E. coli* and antiglobulins to them as ingredients in enzyme immunoassay test systems.

It was found that 5  $\mu$ g/ml for 38 kD protein and 10  $\mu$ g/ml for 18 kD protein were the optimum doses for sensitizing polystyrene trays with antigen. The main advantage of using these proteins in the test system is their high specificity. The system does not demonstrate antibodies to heterologous microorganisms with the same antigenic determinants, such as *Y. enterocolitica* O-9.

Use of specific immunoglobulins for these proteins as sensitizer of the polystyrene trays in a dosage of 10  $\mu$ g/ml enabled us to demonstrate soluble brucellosis antigen (isolated from various species of brucella in the S form) in a dose of 1-5 ng/ml, which is consistent with the resolution capacity of enzyme immunoassay.

In 1994 we plan to continue the study of immunobiological properties of cloned proteins and assess efficacy of diagnostic test systems based on these proteins and immunoglobulins to them.

#### Recovery of Recombinant Strains Producing Human Cytokines

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It has now been shown that cytokines can be used effectively to diagnose and treat a number of diseases. The most widespread source of these substances in sufficient quantities for scientific studies and use in

medicine are artificially developed bacterial strains that produce recombinant cytokines.

In 1993, our studies proceeded in two main directions: development of a strain that produces recombinant protein of interleukin-1 receptor antagonist (hIL-1ra) and cloning of the gene that transforms the human beta growth factor (hTGF- $\beta$ ).

To obtain a strain producing recombinant hIL-1ra we used the gene for interleukin-1 receptor antagonist protein recovered in the laboratory of Yu. A. Berlin (RAS Institute of Bioorganic Chemistry). To achieve effective translation of the hIL-1ra gene we used the technique for designing a hybrid operon with partially overlapping genes (TGATG vector). The yield of recombinant hIL-1ra constituted about 6 percent. The purified protein had the activity of native hIL-1ra.

To clone the TGF- $\beta$  gene, we designed a collection of cDNA synthesized on poly(A)+RNA isolated from human placenta. The collection of cDNA in phage vector  $\lambda$ gt10 contained more than  $1 \times 10^6$  independent recombinant clones. Oligonucleotide probes corresponding to sites in the gene that codes the amino end and C end region of mature protein were synthesized on the basis of published data in order to scan the cDNA collection for TGF- $\beta$ . After several successive screenings, we selected two clones containing foreign DNA fragments about 1200 and 1500 n.p. [nucleotide pairs] in size, which were hybridized with both oligonucleotide probes. We assume that the clones we selected contain cDNA TGF- $\beta$  fragment suitable for creating a strain that produces this protein. In 1994 we plan to continue the work on development of strains that produce recombinant human cytokins, as well as to complete work on development of a strain producing recombinant TGF- $\beta$ , and to study conditions for superproduction of recombinant hIL-1ra and TGF- $\beta$  in *Escherichia coli* cells. We also plan to study the efficiency of production of recombinant human interferon- $\alpha$ -2 by *Escherichia coli* with use of two variants of the gene for this protein in a system of expression that is dependent on RNA polymerase of bacteriophage T7.

#### Cloning of Destabilase Gene From Medicinal Leech and Recovery of Producing Strain

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Destabilase is an enzyme of the medicinal leech that is involved in resorption of thrombi. The purpose of this study was to clone the destabilase gene and recover a producer of this enzyme.

We isolated mRNA from the leech and recovered cDNA on its basis.

On the basis of the partially identified amino acid sequence of destabilase we synthesized degraded oligonucleotide primers. The needed DNA fragments were recovered using these primers in the PCR [polymerase chain reaction] test based on leech cDNA. These fragments were cloned and sequenced. As a result, two different isoforms of destabilase were found: Ds1 and Ds2, the amino acid sequences of which, as predicted on the basis of the nucleotide sequence, had a high degree of homology (75 and 63 percent, respectively) with the amino acid sequence of the enzyme recovered earlier when its primary structure was studied.

In 1994 we plan to recover full size cDNA of the two destabilase isoforms and to develop strains that produce these proteins.

#### Design of a Model *B. Pertussis* Strain Producing the Toxoid Form of PT

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The chief constituent of acellular pertussis vaccine (PV) is the toxoid form of pertussis toxoid (PT), which may be produced by recombinant strains designed on the basis of a heterologous recipient or recovered by substitution of the native PT operon in the *B. pertussis* chromosome with a mutant operon.

The purpose of this study was to assess the expediency of using *B. pertussis* and *Ps. putida* as recipients to recover strains producing a nontoxic immunologically active form of PT (toxoid).

To achieve the set goal, we cloned the PT mutant operon contained in the vector of a wide range of hosts controlled by the *E. coli* lac promoter. The new recombinant pNAS plasmid was transmitted by the electroporation method into cells of an atoxigenic mutant strain of *B. pertussis* 357 tox:Tn5. The quantity of PT produced by recombinant clones was measured by the IFA method. Its toxic properties were identified in experiments on Chinese hamster cells (CHO). The level of toxoid production by the recovered recombinants was up to the level of the native Tohama strain (2.8-3.5  $\mu$ g/ml PT). Toxicity of the mutant PT was lowered by a factor of 10<sup>3</sup>-10<sup>4</sup>. Thus, a strain producing a nontoxic PT derivative was designed on the basis of a nontoxigenic mutant strain of *B. pertussis* 357.

We studied some of the physicochemical and biological properties of the protein produced by the recom-

binant *Ps. putida* strain we designed, which carries a mutant PT operon in the plasmid. The recombinant protein has diminished toxicity for CHO cells (1000 times less), leukocyte-stimulating (4-5-fold) and histamine-sensitizing (to the baseline level) activity. The protective activity of recombinant cell disintegrates in mice infected intracerebrally with virulent strain of *B. pertussis* 18323 constituted 80 percent, which corresponds to the protection provided by lysates of the native vaccine strain of *B. pertussis* 475.

Ultrasound disintegrates were removed from recombinant protein with use of affine column chromatography with immobilized antibodies to native PT. The purified preparation contains proteins similar in molecular weight to subunits of native PT that interact with serum to native PT, as well as a protein with molecular weight of about 60 kD, which does not interact with serum to native Pt. The nature of this high molecular peptide is under investigation.

#### Recovery of Recombinant Epidermis Growth Factor, Study of Its Effect on Cell Growth and Tissue Regeneration

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In order to study the properties of vaccinia virus recombinant growth factor (VEGF), this protein was purified in stages with determination of activity at each stage of isolation. The final purification stages consisted of chromatography on DEAE cellulose and gel filtration on sephadryl. Purity of the preparation was determined using protein electrophoresis in polyacrylamide gel. The recovered protein was identified by the immune blot test using previously obtained antibodies on VEGF.

A study of acute and chronic toxicity using presumptive therapeutic doses and 10 times higher doses revealed the safety of the obtained VEGF preparations.

Biological activity was determined according to binding of recombinant VEGF with specific receptors on the surface of formalin-fixed A-431 cells and in experiments on albino rats for demonstration of burn-treatment effect of this protein. Experiments involving binding of recombinant growth factor with receptors on the surface of A-431 cells revealed that VEGF interacts specifically with receptors at the base and surface of microvilli of A-431 cells.

A comparison of burn-treatment action of VEGF to a standard therapeutic agent (levomecol) revealed that

there is more effective restoration of the destroyed epidermal layer with VEGF treatment than with Levomecol treatment.

In 1994, we plan to recover a producer strain expressed in VEGF in the form of mature protein with a supplement of nine amino acids at the N end, and to study its biological properties.

#### Optimization of Methods for Introducing Plasmid DNA in Propionibacterial Cells

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Propionic acid and vitamin B<sub>12</sub> stored intracellularly, and a few other products of vital functions of propionibacteria of the dairy group are the reasons for interest to biology and genetics in these microorganisms which are used in the food industry and agriculture.

To develop a vector-host system, we optimized the method of accumulating cells of these bacteria and the procedure for isolating plasmid DNA. We studied the capabilities of different methods of introducing plasmids with a wide range of hosts into propionibacterial cells. It was shown that the original vector plasmid pLFR1 [5 t.b.p. [thousand base pairs], determines resistance to chloramphenicol (Cm)] can be introduced into strains of *Propionibacterium freudenreichii* F3 and of *P. acidipropionici* F2 through conjugative mobilization from *Escherichia coli* at frequencies of about 10<sup>-7</sup> scaled to the concentration of recipient cells. Evidently, the level of expression of foreign genes in propionibacterial cells is low. This is manifested by slower growth of recombinant clones under selective conditions and instability of the vector, since introduced plasmid DNA could not be detected electrophoretically in the recombinant preparations.

It was established that electrotransformation under conditions of partial discharge of a memory capacitor provides for introduction of pNZ12 (4.1 t.b.p., Cm) and pSL211 (9.7 t.b.p., determines resistance to erythromycin and lincomycin) plasmids in F2 and F3 strains with effectiveness on the order of 10-2x10<sup>2</sup> transformants/μg transforming DNA when cell specimens are submitted to single electric pulses of a square, half-wave, sinusoid or complex shape within the range of electric field voltage of 13 kV/cm and pulses lasting 1.3- 2.6 ms. Since we used preparations of plasmids isolated from *E. coli* cells, the observed effectiveness of electrotransformation was probably close to optimum. These plasmids were also notable for instability in F2 and F3 strains. Hybridization analysis of recombinants' DNA by the Southern method revealed that the pSL211

plasmid could integrate in the chromosome of appropriate transformants.

Under selective conditions, F3 transformants containing pSL211 DNA are stable, they form colonies of the usual size after 8-12 days of anaerobic incubation. Consequently, the vector elements containing pSL211 plasmid DNA (pCB20 derivative) can be used as integrative vectors for *P. freudenreichii* strain F3.

#### Development of Recombinant Strains Producing Protective Antigens of Pathogenic Species of the Genus *Yersinia*

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The purpose of this work was to design recombinant strains that produce protective antigens of pathogenic bacteria of the genus *Yersinia* (*Y. pestis*, *Y. pseudotuberculosis*, *Y. enterocolitica*). The object of the study was the pCad calcium-dependent plasmid (45-47 MD) common to *yersinia*, the products of which play a leading part in pathogenicity and immunogenicity of these pathogens. This plasmid codes synthesis of outer membrane proteins (OMP or YOPs — *Yersinia* outer membrane proteins), antibodies to which provide passive protection against plague and other *yersinioses*. There is a problem in recovering these polypeptides, since they are synthesized in vitro only under conditions of cellular stasis (temperature of 37°C and shortage of Ca<sup>++</sup> ions), and under such conditions the pCad plasmid is unstable. It is proposed to solve this problem by designing producer strains with recombinant replicons and individual calcium-dependent plasmid genes under the control of *E. coli* promoters.

The purpose of studies carried out in 1993 was to chart structural and functional maps of the fifth HindIII fragment of the *Y. pestis* pCad358 plasmid that codes synthesis of OMP (46 and 31 kD) and polypeptide with mol. wt. 28 kD. As a result, a restriction map was charted of the fifth HindIII fragment for BamHI, BglII, EcoRI, HpaI, KpnI, PstI, PvuI, XbaI and XhoI endonucleases. On this map, the position of three genes coding synthesis of proteins with mol. wt. of 46, 31 and 28 kD was identified. Recombinant plasmids were designed that carry separate genes and different combinations thereof. Expression of recombinant plasmids was studied in vivo and in *E. coli* mini-cells. A comprehensive study was started of the structure of the gene for the 46 kD protein (OMP2), and the site of the promoter of this gene has been tentatively determined.

In 1994, we plan to carry out a comprehensive study, based on our findings, of regulation of expression of

the gene for OMP2 protein, as well as optimize conditions for producing it by selecting a suitable vector, host strain and cultivation conditions, and to design an effective OMP2-producing strain. Recombinants with cloned genes coding synthesis of proteins with mol. wt. of 31 and 28 kD will be used to study protective properties of these polypeptides and to develop producers.

**Cloning of Genes Responsible for Synthesis of Antibiotics With a Broad Spectrum of Action (Microcins), and Design of Bacterial Strains for Treatment of Farm Animals and Man**

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Deletion and insertion mutations of plasmids leading to absence of microcin synthesis and immunity were recovered to study plasmid genes involved in synthesis of microcin C51 with the broadest spectrum of action of all known microcins. By combining in a cell pairs of compatible mutant plasmids containing different vectors, complementation analysis was carried out of *mic* and *imm* mutations. Results of this analysis revealed that at least three plasmid genes are involved in microcin synthesis and two genes in expression of producer cell immunity to it.

New data were obtained on the structure of microcin C51. It was shown that microcin is a nucleoside peptide with mol. wt. of about 1200 kD, and it contains 7 amino acids. Using blot hybridization, a mixture of oligonucleotides was synthesized that correspond to the sequence of these amino acids; a plasmid fragment was identified that contains a site coding microcin peptide.

It was shown that there is homology of plasmid fragments determining type B microcine production from *E. coli* strains of different origin. No homology is observed of *Mic* fragments responsible for synthesis of different types of microcins, B and C. The obtained data are indicative of absence of constituents in common in the genetic systems that determine synthesis of different types of microcins and immunity of producer cells to them.

The technology has been developed for experimental industrial production of a preparation based on the microcin-synthesizing strain of *E. coli* recovered in the laboratory.

**Screening and Design of Bacterial Strains for Stimulation of Plant Growth and Biological Control of Their Diseases**

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It was shown that use of bacterial preparations based on antagonist strains of phytopathogenic bacteria and fungi is effective in protecting plants against diseases caused by these microorganisms. Antagonist bacteria were used with success to protect cabbage against root rot, cucumbers and tomatoes against fusariosis, roses and parsley against powdery mildew.

There was testing of the stimulating effect of isolated pseudomonad strains on growth of wheat seedling roots. Two strains were selected that stimulated root growth appreciably.

Adaptation of two antagonist bacterial strains to cucumber seedlings and carnation plants was studied. It was shown that there was dramatic prevalence of added bacterial cultures over other microorganisms after 20 days of plant growth in the microflora that developed on green cucumber seedlings grown from bacteria-treated seeds. There were considerably fewer antagonist bacteria in soil than on the cucumber seedlings. Good adaptation of antagonist bacteria was observed as well in carnation plants. A study of spontaneous mutants of two *Pseudomonas* antagonist strains of phytopathogens revealed that phenazine pigments play the deciding role in antagonistic activity of these strains. An orange phenazine pigment was isolated from cells of a *Pseudomonas aureofaciens* strain. It was shown that this pigment inhibits growth of several phytopathogenic fungi that are the pathogens of root rot and vascular diseases of plants.

Work has begun on introduction into cells of active antagonists of *Bacillus thuringiensis* genes responsible for synthesis of entomopathogenic toxin in order to design bacterial strains to protect plants not only against phytopathogenic bacteria and fungi, but also insects.

In 1994, it is planned to continue testing the effects on plants with various diseases of agents based on the recovered bacteria that are antagonists of phytopathogenic microorganisms, as well as to continue work on introduction of antagonists (*Pseudomonas*) of *B. thuringiensis* genes responsible for synthesis of entomopathogenic toxin, and to test various genetic systems for introduction of transposons into the cells of the pseudomonad strains studied.

**Project 4. Development of Economically Valuable Plants With Gene Engineering. Transgenic Plants**

**Design of Plants With Exogenously Controlled Expression of Foreign Genes**

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In the course of two successive transformations, transgenic tobacco plants were obtained with genetic elements integrated in the chromosome that included the *nptII* gene under a constitutive *nos* promoter, as well as its nonsense form under inducible promoter *hsp70* of the gene for heat shock protein from *Drosophila melanogaster*. Virtually complete suppression of neomycin phosphotransferase II activity in such plants was observed only in response to heat shock. The same effect was observed in experiments on temporary expression of the *nptII* gene introduced into protoplasts of transformed tobacco containing only the *nptII* nonsense gene under the *hsp70* gene promoter.

Consequently, in transgenic plants the promoter of the gene for heat shock protein can be used in nonsense genetic structures for induced suppression of genetic expression.

Plasmids were designed for transcription of the *nptII* gene in the sense and antisense directions for the purpose of further analysis of antisense inhibition.

In 1994, we plan to work on hybridization of transgenic plant RNA, as well as to recover new genetically antisense structures including phytohormone genes of *Agrobacterium tumefaciens*.

#### **Recovery and Introduction Into Culture of Genetically Transformed Roots of Valuable Medicinal Plants**

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We continued the study of chemistry of secondary metabolites of roots of the medicinal plant *Withania somnifera* cultivated under sterile conditions. As a result of extraction with methyl alcohol followed by silicagel column chromatography of many grown roots, several fractions were isolated in which comparative thin-layer chromatography with methanol extract of whole plant roots revealed steroid substances. Three substances were isolated and purified for subsequent mass spectrometry and identification.

On the basis of the results of mass and NMR spectroscopy the structure of the two substances was identified. They are typical steroids, the structure of which resembles the structure of vitanolids described in the literature.

Concurrently with the chemical study of *W. somnifera* roots, work was continued at the Oncological Center, where studies were carried out of the physiological activity of extracts recovered from cultivated roots. Primary screening of obtained preparations in a culture

of lympholeukemia p388 cells and solid LLC carcinoma of the lung revealed elements in root extracts that had cytotoxic and antineoplastic activity. Specialists in the chemotherapy department of the RAS Oncological Research Center concluded that the extracts of *Withania somnifera* root culture are of interest for the purpose of further investigation of their antineoplastic activity in *in vivo* systems, which requires sufficient funding.

In 1993, genetically transformed roots of *Apocynum cannabinum*, which is a medicinal plant whose roots are the raw material for cardiac glycosides of the strophanthin group, were introduced in a culture. Primary chemical analysis of the obtained root culture revealed presence of zimarin, a derivative of K-strophanthin.

#### **Development of a System for Transformation of Sunflower Plant to Obtain Economically Valuable Plants**

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A system is being developed to transform somatic cells of the sunflower plant, which is based on using runner hypocotyl and apex as explants. The work is being carried out on the Peredovik variety.

Determination was made of conditions for cultivating runner hypocotyl and apex to provide for a higher yield of regenerants. Media containing different combinations of phytohormones were used to recover regenerants from hypocotyls. A medium for runner reproduction was used to cultivate hypocotyl segments in order to augment the yield of regenerants. Its use increased the regenerant yield by several times, as compared to the control. Apices of runners of different ages were also used as explants to recover regenerants. Two explant variants were studied: in one variant the apexes were placed whole on medium, and in the other they were cut lengthwise into two halves.

Since the sunflower cultivar is a complex population formed of several lines, plant reactions to the same medium differed, and this is what we observed in our experiments.

In both explant variants plants were recovered, but the quantity varied. In three variants of medium, about 30 percent of the apexes formed numerous runner rudiments. Regenerants were capable of developing roots and inflorescences.

During cultivation of regenerants recovered from the apical meristem we observed formation of numerous runner primordia on the leaves. Conditions for rooting of multiple regenerants formed on the leaf surface are under study.

In order to obtain stable transformation, in addition to mass regeneration it is necessary to obtain regenerants with maximum viability, since introduction of foreign genes reduces dramatically survival of regenerants.

For this reason, in 1994 we plan to optimize regenerant cultivation conditions to obtain a high degree of viability.

#### Design of Transgenic Plants Expressing Transport Genes to Develop Resistant Cultivars

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It was shown that *N. tabacum* plants expressing the 32K gene for bromegrass mosaic virus (BMV), which is not present in them, manifest a considerable level of resistance to tobacco mosaic virus (TMV), but remain susceptible to potato X virus (PXV) which belongs to taxonomic groups other than BMV and TMV.

The immune blot method was used to analyze lines of *N. benthamiana* plants transformed by structures containing 25K, 12K+8K, 25K+12K+8K genes of the triple block of transport genes (PXV). 25K was found in the form of a monomer and in a higher molecular form (40K) in all 4 lines containing the 25K gene. 12K and 8K proteins could not be demonstrated.

A model system was found for identification of the gene responsible for distant transport (DT) of PXV. The type strain of barley striped mosaic virus (BSMV) with DT deficiency acquired the capacity for systemic infection in the presence of PXV in *N. benthamiana* plants.

The type strain of BSMV was demonstrable in the upper noninoculated leaves of transgenic plants that produce 25K TB triple block genes. These preliminary data lead us to assume that the 25K gene of the triple block of genes of this virus is responsible for PXV DT.

It was also shown that RNA 3 containing the genes for structural protein (SP) and protein 3a is responsible for distant transport of cucumoviruses. Structures of hybrid RNA 3 containing genes 3a and SP of different cucumoviruses were obtained to identify the gene controlling distant transport. In 1994 we plan to determine the role of 12K and 8K proteins in transport function of PXV, as well as to identify the cucumovirus gene (domain) responsible for distant transport, study the role of membrane protein in this process, and to obtain transgenic plants expressing the 8K protein of PXV.

#### Investigation of Molecular Mechanisms of Osmotic Stability of Plants on the Model of Transgenic

#### Plants With Mutant Bacterial Genes for Osmoprotectors

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Work was done to obtain a spontaneous osm mutant for the proB gene of *Escherichia coli*. To produce mutations we used *E. coli* bacteria containing sex factor F104. A mutation was obtained by means of two-stage selection with use of a proline analogue, L-azetidine-2-carboxylic acid (Azc) and high concentrations of NaCl. Mutant cells were described. It was established that mutant cells grow more slowly than nonmutant ones on minimum medium (MM) without NaCl. However, in MM with 0.6 M NaCl, nonmutant cells do not grow, whereas mutant ones grow at the rate of 0.25 generations per hour. The maximum concentration of Azc in which mutant cells grow is 20  $\mu$ g/ml. There was 10 times more proline in mutant cells in MM without NaCl than in nonmutant cells; moreover, up to 150  $\mu$ g/ml proline was excreted into the medium. In MM with NaCl mutant cells did not excrete proline into the medium and contained 30 times more proline than nonmutant cells. Mutant cells stop growing with 0.9 M NaCl, they "feed" pro cells on agar-containing MM and grow on MM with 0.7 percent succharose.

#### Development of Transgenic Plants

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A vector was recovered for expression in plants of the *P. putida* nahC gene under the control of an inducible hsp promoter.

Transgenic tobacco plants were obtained with the beta-1,3-glucanase gene from *Clostridium thermocellum* under the control of rbcS promoters from *arabidopsis* or the 1' gene from *Agrobacterium*. The same strains were used to infect potato explants. As a result, transgenic potato runners were obtained, into which the glc gene was introduced. We continued development of methods for transformation of different potato cultivars, ipt and xyl genes were introduced into Lugovskaya and Golubizna cultivars, and growth of transgenic runners was obtained.

Expression of chloroplast gene RBFK (rbcL) and a few other genes was analyzed in previously developed trans-

genic tobacco plants with the *ipt* gene, with elevated level of endogenous cytokinin. According to results of blot hybridization of RNA, relative accumulation of a transcript of the *rbcL* gene in the total RNA preparation of transgenic plants was 2-3 times greater than in the control. The level of transcription of several other chloroplast genes was also altered.

We plan to analyze expression in transgenic plants of introduced *nahC* and *dic* genes. We will study the effect of *nahC* gene on levels of phenol compounds and activity of plant polyphenol oxidase. We will study the effect of *glc* gene expression on plant resistance to stress. We expect to recover new vectors for controllable expression of the gene for synthesis of cytokinin in transgenic plants.

#### Development of New Vector System on the Basis of Agrobacterium *Tumefaciens* Strain A281

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Tobacco (*N. tabacum*) was transformed by co-cultivation with leaf disk agrobacteria. Helper strain CBE21, the T-DNA of which contains the marker for resistance to hygromycin (*hph*) and binary vector pMON 505::bar, was used for transformation. To obtain a binary vector, the fragment containing the 35S promoter, phosphinotricin acetyltransferase gene (*bar*) and nos terminator were cloned in vector pMON505 (Horsch, Klee, 1986). The pMON505 contained in T-DNA contains the gene for neomycin phosphotransferase II (*npt*) with promoter and terminator nos.

A fragment of the expected length was obtained in all 10 plants when they were tested by PCR with primers specific to the *hph* of these plants obtained on a medium with hygromycin. The appropriate fragment was obtained in 5 of these 10 plants with primers for *bar*. In the same five plants, a positive signal was also obtained upon testing with *npt* primers. Thus, with use of this vector system plants with two T-DNA appear at a high frequency.

As a result of self-pollination plant seeds were obtained containing both T-DNA in their genome. The offspring of one of the original "double" transformants obtained on medium with hygromycin was analyzed using PCR. Among the 32 plants analyzed, there were 3 containing the *bar* gene but not the *hph* gene. Thus, using the CBE21 strain one can obtain transgenic plants without the selective marker of antibiotic resistance provided that the T-DNA of the binary vector used does not contain this marker.

We cloned and sequenced fragments of DNA pTiB0542, in particular a fragment containing gene 6b and the

nontranscribable region preceding it and overlapping T-DNA fragments 441 and 754 n.p. in length containing the right margin of T-DNA transfer. In 1994, we plan to design a binary vector whose T-DNA does not contain the marker of antibiotic resistance using in particular fragments of T-DNA pTiB0542.

#### Design of Transgenic Plants Resistant to Insect Pests

Ye. B. Rukavtsova, I. G. Bogdarina, S. V. Dolgov, and Ya. I. Buryanov, Affiliate of RAS Institute of Bioorganic Chemistry, Pushchino

Analysis was made of insecticidal activity of F0 and F1 generations of transgenic tobacco plants containing in their genome the gene for *Bacillus thuringiensis* var. *berliner* delta endotoxin under the control of promoter I' of the mannopin synthase gene. It was shown that these plants are toxic to gipsy moth larvae, tobacco thrips, spider mite, Colorado beetle larvae, clover and alfalfa cutworms. Transgenic plant seeds were transmitted to the Tobacco Scientific Production Association (Krasnodar) for field tests.

The high toxicity of obtained plants is perhaps attributable to the dramatic increase in toxin synthesis after plants are attacked by insects. It is known that the TR1'-2' promoter is induced after plant cells are injured and, moreover, is regulated by hormone content of specific plant organs. We have shown that expression of the gene for neomycin phosphotransferase II from promoter TR2' increases by many times in injured leaves of transgenic plants. The level of such expression is probably comparable to expression from promoter 35S. Analogous tests are being conducted at the present time with plants containing the gene for delta endotoxin under control of promoter TR1'.

In 1994 we plan to continue experiments for comparative analysis of the gene for delta endotoxin of *Bacillus thuringiensis* var. *berliner* from promoters TR1' and 35S in transgenic tobacco and chrysanthemum plants.

#### Development of Transgenic Potato, Tomato, Soybean and Alfalfa Plants With Improved Features by Introduction of Bacterial Genes

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The leaf disk method was used, with agrobacterial infection of plants in order to develop glyphosate-resistant potato plants (Lugovskoy and Resurs cultivars).

The integrative pPREP3 vector we used contained the mutant *aroA* gene fused with a sequence coding the transit peptide of the small subunit of pea ribulose diphosphate carboxylase (to provide for transport of mutant EPSP synthetase into chloroplasts) under the control of the 35S promoter of cauliflower mosaic virus. The callus lines obtained grew on medium containing up to 3 mM glyphosphate.<sup>2</sup> However, the selected lines of transgenic plants withstood treatment with up to 30 mM glyphosphate, which is 3 times higher than the concentration used in the fields.

In the work done to recover transgenic tomato plants (Bakhtemir cultivars), which express the gene for biosynthesis of cytokinin (*ipt*), three transformants each were selected from breeding lines 101 and 108. Transgenicity of the tomato plants was confirmed using the PCR method and NP-TII activity. Assay of phytohormones and analysis of expression of some stress genes in the obtained lines confirmed the higher (by about 1.5-2 times) level of zeatins. There was higher peroxidase activity in the transgenic plants (particularly the roots). We succeeded in recovering seeds as a result of self-pollination for one of the transgenic plants. Seed yield was 35-50 from 3 tomatoes. Seed germination has not yet been determined.

In the work dealing with recovery of transgenic clover and alfalfa plants, cuttings of 7-day seedlings constituted the most effective recipient system. The process of co-cultivation of agrobacterial strain pGLTr4 (which contains the gene for biosynthesis of cytokinin *ipt*) with explants took place at the site of cutting the hypocotyl. As a result, growth of dark-green teratomic structures (in the case of alfalfa) from the upper part of the cutting and buds from the lower was observed on hormone-free medium. In the case of clover, the callus formed on cuttings of the upper part of seedlings differed from the lower part in that there was faster growth and rhizogenesis. The obtained plants — clover and alfalfa regenerants — grew on medium with kanamycin. Addition of exogenous hormone to the medium suppressed the regeneration process entirely.

#### **Recovery of Transgenic Clover Plants (*Trifolium Pratense*) Resistant to the Herbicide Phosphinotricin (BASTA)**

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Several clones of regenerant plants recovered from explants of hypocotyles from seedlings of the VIK7 cultivar developed by the Russian Feed Institute were selected for genetic transformation of clover (*Trifolium*

*pratense*). The clones were notable for regenerative capacity.

Two types of structures were observed in segments of regenerant plant petioles placed on medium for callusogenesis: friable light callus tissue and neoplastic growth. When explants were transferred to medium to induce morphogenesis, the first type showed proliferation of the callus, which turned green and subsequent runner formation. The second type was characterized by appearance of solid green globules at the ends, from which runners and roots developed.

Agrobacterium tumefaciens strain A281 bearing "disarmed" plasmid pTi-Bo542 with the plant selective marker of resistance to hygromycin and kanamycin and chimeric phosphinotricin acetyltransferase (bar) gene, which determines resistance to the herbicide phosphinotricin (BASTA).

Transformation was effected by co-cultivation of agrobacteria and explants of plant petioles *in vitro*. Inoculated explants were put in selective callus-forming medium with 50 mg/l kanamycin and 500 mg/l claforan. In such a medium, the first type of clones formed a kanamycin-resistant callus, whereas neoplastic growth was observed in clones of the second type. When the formed structures were transferred to medium for morphogenesis, the first type of clones did not form morphogenic structures, while the second type regenerated runners, which remained green and rooted in selective medium. Control plants did not take root in selective medium. Control plants did not take root with kanamycin in a concentration of 5 mg/l. Presence of the inserted bar gene in the runners recovered after transformation was proved using the PCR method.

At the present time, transgenic plants have been planted in soil for subsequent analysis of offspring.

#### **Cloning, Expression, Identification of Product and Use of the RSF1010 Plasmid That Suppresses Agrobacterial Oncogenicity to Design Plants Resistant to Bacterial Gall**

L. S. Chernin, L. A. Gukova, T. S. Denisova, I. Kh. Kushner, and M. A. Terentyev, RAS Bioengineering Center, and All-Russian Research Institute of Agricultural Biotechnology of the Russian Academy of Agriculture, Moscow

Sixteen kanamycin-resistant tobacco transformants were recovered that contain the *ita* locus of plasmid RSF1010 in vector pPCVRS-75. The presence of sequences of the hybrid vector plasmid with an RSF1010 fragment was confirmed by PCR analysis. Half of these transformants did not develop tumors after the first test for resistance

to gall, another six developed tumors after 2-3 repeated inoculations. The remaining plants were resistant to bacterial gall.

The vector system used at this stage of our work did not enable us to check for expression of the ita locus in bacterial and plant cells. To design a vector with verifiable expression at all stages of cloning and transformation the ita locus was recloned in two directions in an expression vector cassette with pRT101 vector under the control of the 35S promoter. Then the obtained expression vectors with the ita locus in different directions were cloned in a set of binary series pBIG vectors. This is of basic importance for ongoing monitoring of antioncogenic activity of the ita locus. The obtained series of four types of structures was transmitted through three-parent crosses to *Agrobacterium tumefaciens* strain LBA4404. Tobacco plants were transformed by the leaf disk method with a set of obtained strains bearing hybrid plasmids based on binary vectors. The recovered iDi strains, which contain the same set of structures, are undergoing oncogenicity tests.

A model system including lines of plants with imbalance in level of phytohormone production was developed to test the capacity of the ita gene product to inhibit expression of onc genes in plants infected with agrobacteria. Sensitivity of plants in this model system to bacterial gall was analyzed.

We plan to test new transformations for resistance to crown gall, as well as to carry out a molecular genetic study of transformants with the ita locus, its expression in plant and bacterial cells, and to study its inheritance in transformant progeny.

#### **Development of Transgenic Potato Plants Expressing Bacterial Genes That Affect the Plant Polyphenol Oxidase System**

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In 1993 studies were carried out and the following results were obtained:

1. The effect of lowering cultivation temperature for transformed explants, as well as addition to nutrient medium of ascorbic acid as antioxidant were studied in order to optimize conditions for transforming potato cultivars with low polyphenol oxidase activity. Both procedures diminished the number of explants that darkened and became necrotic.

2. Plants were recovered from transformed explants that were evaluated for effectiveness of callusogenesis in the presence of 25 mg/l kanamycin. One-third of the regenerants demonstrated active formation of callus tissue.

3. Transformants of two potato cultivars, Filatovskiy and Lugovskiy, were retested for polyphenol oxidase activity in order to confirm expression of the nahC gene.

4. A vector was developed on the basis of the hsp promoter for thermoinducible expression in plants, and the nahC gene was cloned in it. As a result, an agrobacterial strain was recovered for transfer into plants of the induced variant of the nahC gene.

In 1994, we plan to carry out transformation with use of a thermoinducible vector, as well as to obtain transgenic plants, assess expression of the nahC gene, and determine polyphenol oxidase activity in transformed plants.

#### **Isolation of Genes for Resistance to Herbicides, and Development of Herbicide-Resistant Transgenic Plants**

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Work has been completed on cloning, sequencing and mutation analysis of a new gene responsible for resistance to the herbicide difunon, which inhibits synthesis of carotenoids in cyanobacteria and plants. The dfr gene codes protein (74 kD) which has a domain of protein kinase activity inherent in sensory proteins involved in phosphorus transport and metabolism. Insertions in the gene also elicit development of resistance to chlorproazine that inhibits  $Ca^{++}$ -dependent proteins, which is indicative of possible involvement of the gene product in adaptive response systems related to the effects of calmodulinoid proteins. This is a new, previously unstudied mechanism of development of resistance to herbicides of the difunon type.

A clone the partial sequencing of which revealed homology with the cyanobacterial gene was isolated with use of the cyanobacterial dfr gene as a probe from the collection of tobacco cDNA. This is indicative of presence in the plant of a gene the function of which may be related to the difunon-resistance trait.

A collection was formed of cyanobacterial mutants resistant to the herbicides paraquat and amitrole, which inhibit different stages of electron transport and elicit oxidative stress in plant cells. Genome banks were created from two types of paraquat-resistant mutants and two independent amitrole-resistant mutants.

### Recovery of Transgenic Tobacco and Potato Plants Resistant to the Herbicide Phosphinotricin

O. A. Shulga, L. S. Padegimas, and K. G. Skryabin, RAS Bioengineering Center, Moscow

Work was done to obtain transgenic tobacco and potato plants resistant to phosphinotricin. For this purpose, the native gene of phosphinotricin acetyltransferase (pat gene) was isolated from a fragment of *Streptomyces hygroscopicus* genomic DNA cloned at the Research Institute of Genetics of Industrial Microorganisms, and using PCR the GTG-initiating native clone was replaced with ATG. The obtained fragment, which contained the pat gene with altered initiating codon was cloned in M13 and 200 nucleotides were scanned at each end using the Sanger's sequencing method. It was shown that all of the changes occurred as planned, whereas the sequence of the rest of the pat gene region coincides with the one described previously. In order to obtain expression of the pat gene in plant cells, this enzyme was transferred under control of the 35S promoter of the CaMv and NOS terminator. For this purpose, the GUS gene in vector pB1121 was replaced with the sequence of the modified pat gene. This mount of expression of the pat gene was transferred to *Agrobacterium tumefaciens* strain LBA4404, and the obtained strain was used to transform tobacco (*Nicotiana tabacum* cv. SRI) and potato (*Solanum tuberosum*, Prigozhii-2 cultivar, which is resistant to nematodes) plants. The leaf disk transformation method (Horsh et al, 1985) was used to transfer the pat gene to tobacco plants. To transfer the pat gene to potato plants, stalk segments were submitted to transformation by agrobacteria (Newell et al, 1991), since efforts to transform leaf disks of this potato variety failed with use of the method described by deBloque. These plants are being analyzed at present.

In 1994, we plan to carry out molecular biological analysis of recovered regenerants and to check resistance of transgenic plants to treatment with the herbicide phosphinotricin.

### Adaptation of the Structure of the Gene of Bacterial Insecticide Protein for Expression in Transgenic Plants

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The purpose of this work was to selectively change the structure of the gene for the bacterial insecticide protein delta endotoxin from the entomopathogenic bacterium *Bacillus thuringiensis* ssp. *berliner* in order to adapt it for expression in plants and development of plants resistant to Lepidoptera pests.

Using the polymerase chain reaction with modified primers, we obtained two genetic fragments overlapping the site of the bacterial gene coding the active protein segment. The structure of these fragments contains changes that we assume will aid in effective expression of insecticide protein in the plant environment, namely: 1) extraneous ATG columns which induce false initiation of translation are removed and 2) a stop codon is inserted at the end of the segment being translated.

Preparatory work was done for intermediate cloning of the recovered fragments. In particular, a technique was refined for transformation of *Escherichia coli* strains deficient in recombination activity, and we selected conditions for joining amplified fragments to a plasmid vector.

In 1994, we plan: 1) to complete experiments on joining fragments and their intermediate cloning; 2) to combine cloned fragments in a vector containing controlling sequences inherent in plant genomes; 3) to describe the recovered genetic structure from the standpoint of conformity of its nucleotide sequence to the expected one; 4) to start experiments on transfer of recovered genetic structure into the *Ty* plasmid of *Agrobacterium tumefaciens*. Subsequently, we plan to carry out experiments on transformation and economic value of plants. The funding needed to carry out the work planned for 1994 amounts to 7 million rubles in January 1994 prices.

### Project 5. Development of Economically Valuable Animals Using Gene and Embryo Engineering

#### Transgenic Animals

##### Development of Methodological Approaches to Demonstration of Transgene at Different Stages of Embryogenesis, and Evaluation of Effectiveness of Its Expression

L. Ye. Andreyeva and G. A. Dvoryanchikov, RAS Institute of Molecular Genetics, and All-Russian Research Institute of Physiology and Biochemistry of Livestock Nutrition, Russian Academy of Agricultural Sciences, Moscow

Several series of microinjections of the RSV-lacZ gene were performed into fertilized roe of *Missgurnus fossilis* L. loach. In one of the series of experiments, the gene was injected in the following variants and stages of development: 1) into the cytoplasm of the early blastodisk 10-12 min after fertilization (a.f.); 2) into the vegetative part of the yolk 30-40 min a.f.; 3) into the cytoplasm of the late blastodisk 50-60 min a.f.; 4) into the cytoplasm of one of the blastomeres of the 2-blastomere embryo 70-80 min a.f. Embryos in the

3d and 5th days of development were stained with X-Gal substrate. Expression of transgene in the form of separate blue-green dots, dashes and spots on the surface of the head, body and fins was observed in 100, 30, 90 and 70 percent of 3-day embryos and in 100, 100, 61 and 90 percent of 5-day embryos (10 specimens in each variant), respectively. In another series of experiments the gene was injected in two variants: into the blastodisk cytoplasm and in the vegetative part of the ovicell yolk (10-60 min a.f.). Transgene expression was observed in 100 percent of the 3-day embryos with both variants of injections. DNA was isolated from 5-6-week larvae in both series of experiments. PCT analysis of DNA revealed that the transgene is present in 100 percent of the specimens after injection into the blastodisk cytoplasm and 65 percent of those after injection into the yolk. After *n* lot hybridization of DNA with the RSV-lacZ gene, it was found that the transgene is present in high molecular fractions of loach genomic DNA, and the dimensions of hybridized restricted fragments of genomic DNA were as expected.

A simple technique is proposed to master the procedures used during manipulations of ovicells and microinjections of genes into pronuclei of zygotes and nuclei of somatic cells of mammals. It is planned to use mouse oocytes at the blastocyst stage as object of investigation and *E. coli*

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galactosidase as injected material. A solution of this galactosidase is injected in the blastocyst, which is morphologically similar to zygote pronuclei, but is larger. The oocytes are then fixed in 0.25 percent glutaraldehyde and stained with X-Gal substrate for 2-15 h. Green coloration of only the blastocyst is indicative of accurate injection of solution into the nucleus and preservation of integrity of nuclear and egg membranes of the ovicell.

#### Development of Livestock With Unnatural Form of Resistance to Viral Infections Based on Nonsense RNA and Ribozymes Against Bovine Leukemia Virus (BLV)

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Anti-BLV acRNA genes and corresponding plasmids under the control of VAI promoter cloned from adenovirus hb DNA were designed. DNA fragments were

cloned in reverse direction under the control of the above-mentioned promoter from the three functionally important zones of the BLV genome, regions R, R-U5 and 5' of the untranslatable region of the gag gene. The recovered acRNA genes contained in plasmid pUC184 were tested in an *in vitro* model system of CC81 cells. The reaction of syncytium production, the plasmid with the acRNA gene opposite the R-U5 region of the BLV genome revealed 50 percent inhibition of viral reproduction with a 10:1 weight ratio to the BLV genome. The rest of the structures were less active.

A ribozyme was synthesized for the R region that effectively splits viral RNA *in vitro*.

Work was done to isolate and clone the promoter region of the BLV genome. It was shown that effective expression of genes placed under the control of this promoter is possible only in the presence of specific transactivators during viral infection. On the basis of structures containing reporter  $\beta$  galactosidase genes and secreted alkaline phosphatase under the control of the viral promoter, systems were developed to test activity of acRNA genes and ribozymes in FLK-BLV and CC81 cell lines.

In 1994 we plan to develop plasmids containing several acRNA genes (including ribozymes) and to test their antiviral activity in cell cultures.

#### Design of Vectors Based on Regulatory Elements of Casein Genes, and Study of Expression of Hybrid Genes in Transgenic Animals

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Two variants of vectors were designed that contain segments of the promoter region of the bovine

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casein gene, joined to the structural part of bovine growth hormone (bGH). The vectors also contained segments of signals of splicing and polyadenylation of

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casein and

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casein genes. In order to test the effect of regulatory sequences of the

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casein gene on expression of the bGH gene in mammary gland tissues, both structures were injected into zygotes of (CBAxC57Bl/6)F1 mice. PCR amplification after two independent isolations of DNA from 55 offspring revealed 6 transgenic animals (5 females and 1 male). Offspring was obtained from four females. It was shown that three of them transmitted the transgene to the next generation. Bovine somatotropin content in the milk of nursing transgenic females was assayed by enzyme immunoassay. The level of bGH expression in the milk of one of the specimens constituted 7.5 ng/ml. RNA was isolated from the mammary glands of 5 transgenic nursing mice. We plan to study transgene expression on the transcription level by the RNA amplification method and Nozern hybridization, as well as to perform histological analysis of mammary glands in order to demonstrate bGH by a histochemical method.

**Use of ES Cells to Obtain Transgenic Animals: Search for and Study of Genes Involved in Control of Individual Development**

O. A. Larionov, RAS Institute of Bioorganic Chemistry imeni M. M. Shemyakin and Yu. A. Ovchinnikov, Moscow

In order to obtain a trap vector we used canonical SA and the polyadenylation signal of the small t-antigen of SV40 virus. As sources of lacZ and neo genes we used pVR278 and pSV2neo plasmids, respectively. DNA fragments containing lacZ and neo genes were amplified. A fragment containing the neo gene in XbaI and HindIII sites was cloned in a polylinker of plasmid pUC19. Several clones were obtained (in particular pUC19-neo6) containing the plasmid in which the neo gene was inserted. In the future we plan to clone the DNA fragment containing the lacZ gene in plasmid pBPUC318. Then, the DNA fragment containing the neo gene in ApaI and HindIII sites from plasmid pUC19neob will be inserted in the obtained plasmid. Experiments were carried out to refine conditions for cultivation of mouse embryo stem cells (ES cells).

We also refined the stage of recovery of injection chimeras. Chimeric nature of obtained embryos was determined according to absence or presence of mosaic

pattern in retinal pigmented epithelia of the embryonic eye on histological sections.

**Study of Expression of the Human Gamma-Interferon Gene Under Control of Regulatory Regions of the Ovine Beta-Lactoglobulin Gene in Transgenic Mammals**

O. A. Larionov, RAS Institute of Bioorganic Chemistry imeni M. M. Shemyakin and Yu. A. Ovchinnikov, Moscow

Work was continued on development of transgenic mice expressing human gamma-interferon (hIFN- $\gamma$ ) in the mammary gland. We designed two hybrid genes consisting of regulatory regions of the ovine beta-lactoglobulin gene and structural sequences of the genomic hIFN- $\gamma$  gene. Both constructs (individually) were microinjected into pronuclei of mouse zygotes. Microinjected zygotes were transplanted into the oviduct of pseudopregnant females. Ten transgenic mice (1 male and 9 female) were obtained with the first construct. Thus far only one transgenic mouse was obtained with the second construct.

Transgenic females containing the first construct in their genome were crossed with nontransgenic males. Milk samples were taken from these females on the 12th-14th postpartum day and they were tested for interferon activity. Such activity was found in the milk of all females. In some of them, interferon activity of milk reached 400 ng/ml. In addition, it was shown that the capacity to synthesize biologically active hIFN- $\gamma$  in the mammary gland of female No 14 is inherited by her progeny.

**Development of Vector for Expression of Human Gamma-Interferon in the Mammary Gland of Transgenic Animals**

O. A. Larionov, RAS Institute of Bioorganic Chemistry imeni M. M. Shemyakin and Yu. A. Ovchinnikov, Moscow

A hybrid gene consisting of 5'-flanking sequences of the ovine beta-lactoglobulin (BLG) gene and genomic gene hIFN- $\gamma$ , was designed in order to obtain a vector for expression of human gamma-interferon (hIFN- $\gamma$ ); the AsuII-BamHII fragment of the hybrid gene containing part of the penultimate exon, last intron and part of the last exon of the hIFN- $\gamma$  gene was replaced by the AsuII-BAMHII fragment of another hybrid gene developed previously (this fragment contained the missing part of the last exon of the hIFN- $\gamma$  gene and some sequences of the BLG gene, including its last intron and poly(A) site).

The hybrid gene we developed was used in experiments to obtain transgenic mice.

**Evaluation of Oncological Risk of Mammalian Transgenesis I. A. Serova, Siberian Department of**

**RAS Institute of Cytology and Genetics,  
Novosibirsk**

Analysis was made of long-term sequelae in offspring obtained after manipulating C57BL and F1(BALB<sub>x</sub>DD) mouse zygotes transplanted in syngenic or allogenic recipients. Special attention was given to the study of oncological characteristics of the animals, including morphological analysis of detected tumors. C57BL mice (290 specimens were studied) showed a 1.5-2-fold increase in incidence of tumors regardless of the nature of manipulations (puncture of pronucleus, addition of buffer or DNA). These parameters were not affected by gestation of offspring of the mother-fetus system. F1(BALB<sub>x</sub>DD) hybrids (180 animals studied) revealed considerable, 3-5-fold, increase in incidence of tumors after administration of DNA or buffer, as compared to the intact control.

On the basis of the work done, it can be concluded unequivocally that there is a high potential for injury in the set of methods used to obtain transgenic animals, chimeras, and other embryological work requiring manipulation of zygotes. We previously discovered a high percentage of teratogenic complications in offspring. As can be seen from analysis of long-term sequelae, there is a high incidence of oncological pathology in offspring, particularly of the hybrid genotype.

Thus far we have been unable to single out the stages in early development of embryos that are the most sensitive to mechanical factors. We plan to continue such experiments.

**Recovery and Cultivation of Pluripotent Embryonic Stem Cells (ES) From Mink Blastocysts**

O. L. Serov and M. A. Sukoyan, Siberian Department of RAS Institute of cytology and Genetics, Novosibirsk

ES were used to obtain chimeric mink, and determination was made of dynamics of sister chromatid exchanges (SCE) in ES, in undifferentiated and differentiated condition.

Mink ES were introduced into the mink blastocyst cavity using a microneedle, after which the operated blastocysts were implanted in pseudopregnant females.

A total of 163 recipient blastocysts were extracted from 33 females. MES-12 and MES-16 (both 30,XV) cells were used for injections. These cell lines were selected from 9 obtained in 1992 after evaluating pluripotency under in vivo and in vitro conditions. It was shown that cells of these lines have the capacity to: 1) form multilayer embryonic corpuscles under both in vitro differentiation conditions and after intrauterine injection to nude-line mice (with suppressed immunity); 2) form derivatives of three germinal layers which is graphically illustrated on sections of teratoma-type tumors formed at the sites of subcutaneous injection of ES of MES-12 and MES-16 lines to nude-line mice.

A total of 89 pups were born to 17 "adoptive" mothers, and 32 of them (26 live and 6 stillborn) originated from implanted blastocysts; 44 pups were their own (since not only pseudopregnant but really pregnant females were used in the experiment), while the origin of 13 pups is unclear, since some females ingested their droppings. Thus, blastocyst acceptance was somewhat greater than 20 percent, i.e., quite satisfactory for such transplantation. However, inspection of the pups' coat color revealed that there were no chimeras (chimeric pups would have had mottled coloration).

There may be at least two reasons for absence of chimeras: 1) ES cells used in the experiment have poor tolerance to injection conditions and are not viable within blastocysts and/or 2) injected cells cannot combine with the internal cell mass. In order to determine the true cause, the following work was planned: 1) to inject labeled ES in blastocysts, which would permit tracking their subsequent fate; 2) histological analysis of early stages of mink embryo development. Embryos were extracted from 10 females at different terms of the gestation period, fixed, and the first sections have been made. Histological analysis of these sections, the results of which are tentative, confirm our assumption that ES cannot penetrate into the internal cell mass of the embryo, since the visceral endoderm cells form a dense layer around the presumptive ectoderm.

In 1993, work was started to select a label for ES, and we plan to complete it in 1994.

**Study of SCE [sister chromatid exchanges] in mink ES:** A number of researchers relate the SCE phenomenon to DNA methylation processes. Several studies dealing with early development revealed that SCE level in preimplantation embryos is 2-4 times higher than at later stages of development. We used SCE to study early stages of differentiation of the pluripotent mink stem cells we obtained. Two experimental models based on use of germ cells were used to study SCE during in vitro differentiation: the first consists of pluripotent mink ES capable of forming genuine teratomas and embryoid corpuscles after subcutaneous and intraperitoneal injection to nude-line mice; the second — teratocarcinoma of OTF9-63 mice, the cells of which are capable of differentiation into parietal and visceral endoderm when cultivated with retinoic acid. SCE were counted in undifferentiated cultures, as well as in vitro differentiated cultures at different times after the start of induction of differentiation. We observed a brief elevation of SCE level during spontaneous (mink ES) and retinoic acid-induced (mouse teratocarcinoma) differentiation. The elevation was manifested by 1.5- 2-fold increase in mean number of SCE per metaphase and appearance of cells with many SCE (more than 30) per metaphase. The brief increase coincides with 2-3 cell division in OTF9-63 cells and 3-4 cell division of ES after the start of differentiation. Then the SCE level returns to its initial level inherent in undifferentiated cells. The brief change in SCE level could be the first sign of start of differen-

tiation, which precedes its visible manifestations and, consequently, could serve as a useful indicator of differentiation processes.

#### Footnotes

1. Conclusion. See No 4, 1994 for beginning.
2. Translator's note: Text mentions both "glyphosate" and "glyphosphate" and the terms are rendered as given in source.

#### Eye Injuries Caused by Gas Weapons

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[Article by Prof. R.A. Gundorova, Doctor of Medical Sciences G.G. Bordyugova, and graduate student G.R. Lekishvili, Department of Traumatology, Reconstructive Surgery, and Eye Prosthetics (Prof. R.A. Gundorova, head) of the Moscow Scientific Research Institute of Eye Diseases imeni Gelmgolts (Doctor of Medical Sciences A.M. Yuzhakov, director) of the RF Ministry of Health Care and Medical Industry]

[FBIS Translated Text] At present, companies specializing in the production of personal weapons and self-defense devices produce and deliver each year hundreds of items, which are in great demand ("electroshock," aerosol pistols, gas revolvers and balloons, and clubs). This can be attributed to rising crime rates around the world and the low cost, simplicity of operation, and high efficiency of gas weapons, as well as tight restrictions on the purchase and storage of firearms in many countries, but relatively few restrictions on gas weapons, which recently have been permitted in our country.

In recent years, irritant agents have become most widespread as means of individual protection, while natural compounds, so-called tear gases, or lacrimators, have been used less frequently. Toxicologists do not include irritant agents in the group of chemical warfare agents, but, owing to their specific properties, assign them to an independent group called "police toxic gas agents" in the West. These agents, on contact with human skin, mucosa of the eye, nasopharynx, and upper respiratory tracts, cause injuries to the skin, mucous membranes, and lungs, acute pain sensations, and profuse lacrimation. Quite high concentrations of these agents (on the order of  $10^3$  mg/l) can cause a temporary loss of vision and difficulty in breathing. At higher concentrations, more severe injuries are also possible. When CS and CN get on the skin, redness and blisters may appear.

The irritant effect of lacrimators is connected basically with the presence of halogens in the substance molecule. It increases from chlorine to iodine.

Si-ES gas, or orthochlorobenzenemalonic acid C-dinitrile, is most widespread at present. The colorless

solid substance with melting temperature of 25°C and boiling temperature of 315°C at 760 mm Hg dissolves poorly in water and well in acetone, dioxane, methylene-chloride, ethyl acetate, and benzene.

During World War I about 25 lacrimate substances were tested under combat conditions. Cornea injuries by tear gases were known as far back as that period. Corneal opacities occurred when gas grenades exploded nearby, and the question whether the changes in the cornea should be attributed to the effect of the gas was debated [6]. The first report of a burn to the cornea by chloroacetophenone belongs to W. McNally [14], who reported on three accidents after the use of an apparatus for firing tear gas. After exposure to chloroacetophenone the cornea at first remained transparent, but after a few days an ulcer developed, which led to persistent opacity and vascularization of the cornea [14]. Eye burns caused by close shots from the gas apparatus were often observed subsequently [7, 18]. At the same time, K. Hartmann [4] first drew attention to chloroacetophenone as a harmful substance for the cornea. W. Hopping, P. Laibson and J. Oconor, and H. Tiburtius [5, 9, 17] reported on 40 patients and, among other injuries, described the effect on eyes—reduction in cornea sensitivity, neuroparalytic ulcers, and iridocyclites with secondary glaucoma.

L. Oaks and J. Dorman [15] described two cases of eye burns with subsequent enucleation of one eye. Subsequent reports belong to D. Hoffman, P. Laibson and J. Oconor, R.A. Levine and C.J. Stahl, and R.A. Levine and K. Davidson [7, 9-11].

In the middle of the 1960's gas weapons were represented mainly in the form of sprays and balloons with compressed gas, in which various extracts, mustard oil, and bromacetone were added. M.J. Borer and L.D. Stewart [2] reported on cornea injury as a result of a burn with tear gas owing to the spraying of chloroacetophenone and A. Oksala and L. Salminen [16], after injury with a spray from a short distance, found cornea erosions, edema of the stroma, and folds of Descemet's membrane.

H. Bleckmann and Ch. Sommer [1] reported on 45 patients who were treated in a hospital after burns from tear gas in 1972-1978. In 40 cases cornea burns were the consequence of a pistol shot and in five, of the effect of a spray. One-half of the patients had an erosion of the cornea. During a shot from a gas pistol at a small distance, cavities in the tissue of the cornea were observed in almost one-third of the examined eyes.

The authors also noted stromal edema of the cornea, surface necroses, and folds of Descemet's membrane. Six months after a burn caused by a shot from a gas pistol the central ulcer of the cornea healed with the formation of dense stromal opacity with peripheral vascularization. Among extraocular changes hematomas in the palpebral region and skin burns of a varying degree were noted only in patients with injuries caused

by a pistol shot. In more than 75 percent of the cases during injuries resulting from a pistol shot conjunctival injection was observed and in more than 50 percent, chemosis. In patients injured with a spray, conjunctival injection of the second eye was also observed, and chemosis in all cases. In cases of burns caused by a pistol shot, significant contusion changes in the form of recession of the angle of the anterior chamber, iridodialysis, and traumatic cataract were detected.

H. Bleckmann and Ch. Sommer [1] also warn about the danger inherent in tear gas owing to changes in the cornea, in which in 28 out of 80 eyes injured by a shot and in four out of 10 sprayed eyes a prolonged impairment of visual acuity was noted. After a burn from a pistol, despite intensive treatment, the eyes of one patient had to be enucleated. The authors conclude that a more severe injury occurs with the use of a gas pistol than with a spray [1].

The mechanical trauma from a pistol shot and pressure at the point of impact constitute 200 tech. atm., which is incomparably higher than from a spray, which releases the active substance with a force of about 5 atm.

The question whether it is possible to establish a direct link between the amount of released chloroacetophenone and subsequent cornea injury was studied in experimental and clinical work [1, 3, 12, 13].

Along with corneal toxicity, R.A. Levine and C.J. Stahl [10] described individual sequelae, which made it necessary to remove the eyes 15 years after injury by tear gas. In this connection individual sequelae are attributed to the neurotoxicity of chloroacetophenone, which is manifested in the form of neuroparalytic ulcerations of the cornea. W. Doden and R. Marquard [8] described such a patient with ulcerate necrosis of the cornea after a burn, which did not heal even eight years after the injury.

H. Bleckmann and Ch. Sommer [1] described a case of enucleation of the eyeball three years after an injury caused by a gas pistol shot. Secondary glaucoma, after repeated keratoplasty with a subsequent rejection of transplants and amaurosis, was the reason for this.

Aerosol or gas balloons and manual weapons have now become most widespread as methods of delivery of irritant agents to a target. It is customary to call these weapons "gas pistols and revolvers," although the correctness of such a name raises doubts, because the active substance used in this ammunition is not gas.

Owing to their relatively low cost, simplicity of design, efficiency, and reusability, aerosol balloons with irritant agents are most widespread. The annual production of individual models of such balloons in the West reaches hundreds of thousands. Germany, the United States,

and France are the principal countries producing gas balloons or sprays as means of individual protection.

The aerosol balloon is an aluminum container, as a rule, with a capacity ranging from 20 to 75 ml, filled with a mixture of some solvent (it can be alcohol, benzene, acetone, organochlorine, or ketones) and an active substance, one of the irritant agents. Depending on the concentration of the active substance and its degree of purity, the balloon can contain either a colorless liquid, or a liquid of a yellowish to brown color. As a propellant substance, Freon-11 (CCI 3 F) and Freon-12 (CCI 2 F) usually are used in the balloon. Balloons of some models have safety devices to prevent accidental pressure on the spray head, clips that make it easy to carry them on a belt, or a plastic case for holding them in the hand.

The percentage of the active substance in the mixture contained in aerosol balloons has begun to increase recently. For example, in individual models it comprises 15 to 20 percent, whereas only several years ago, the mixture contained 0.5 to 1.0 percent of the irritant agent.

As a rule, the pistols and revolvers used for gas cartridge firing are exact copies of already available rifled short firearms. The number, name of the model, caliber, and other markings are stamped on every model of a gas weapon in the appropriate places.

Gas cartridges are filled with crystalline chlorobenzylidenemalonitrile powder (CS or SI-ES), or chloroacetophenone. Therefore, in addition to the caliber marking, the manufacturing firm (Geco, Sax, PWS), as well as the active substance used, is indicated on the base of the cartridge case head.

Gas pistols are automatic weapons, because reloading in them and preparation for the next shot are carried out through the effect of powder gases after the first shot. The ejection during a shot of the crystalline active substance at a distance of up to 3 meters is the function of gas cartridges. Cartridges have a cylindrical form and are made from copper and brass.

Caliber 22-Lang cartridges (5.6 and 6 mm—long) are rim-fire cartridges. They do not have a primer in the center of the head base. The percussion composition is located at the head rim and during a shot the weapon striker strikes not in the center of such cartridges, but at their rim, igniting the percussion composition.

Other cartridges of caliber 8 mm PA, 35.45 Short are center-fire cartridges. In the center of the head base there is a primer and, when it is struck by a striker, ignition of the powder charge occurs.

Gas cartridge case mouths are filled with paraffin (22-Lang 6 mm), are rolled in the form of a star (9 mm, 45 Short), or have a plastic plug. Plastic plugs in gas cartridges have different colors: If a cartridge is filled with CS, yellow, and, if the cartridge is filled with CN, blue, violet, or white (depending on the concentration). During a shot, plastic plugs do not fly out together with the active substance, but separate into four petals, and remain within the mouth.

Several reports on eye injuries by gas weapons have appeared in our country's literature in the last few years. However, they are limited to a simple mention of the number of such injuries.

In the Department of Traumatology of the Moscow Scientific Research Institute of Eye Diseases imeni Gelmgolts under Prof. R.A. Gundorova's guidance an experimental-clinical investigation of the changes in the organ of vision during eye injuries by gas weapons is conducted and the first results of experiments on human cadaver eyes, which are of considerable scientific and practical interests, have been published.

Such is the basic information on eye injuries by gas weapons in contemporary ophthalmological literature.

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#### Gene-Based Identification of *Bac. Anthracis* and Soil Saprophytes by the Polymerase Chain Reaction Method

957A0799A Moscow VETERINARIYA in Russian  
May 95 No 5, (signed to press 17 Apr 95) pp 30-31

[Article by T. Kh. Faizov, A. K. Galiullin, and A. M. Alimov, All-Russian Vitamin Research Institute, under the rubric "Infectious Diseases"; UDC 579.842.23.579.8]

[FBIS Translated Text] Current taxonomy and systematics of bacteria are based on an aggregate of diverse phenotypic and genotypic traits. The latter are demonstrated by cultural-morphological, biochemical, and serological methods, as well as modern methods of reassociation, hybridization and restriction of DNA. However, none of these methods provides a complete description of the microorganism genome. The main criterion is genetic identity of specimens that make it up — populations, which is obtained and supported by any of the means of exchange of genetic information (A. B. Yablokov, A. G. Yusufov, 1989).

Currently, multilocus and monolocus genome dactyloscopy methods permit analysis of the genome in its entirety. The latter method permits replication of specific loci of bacterial DNA, in particular identification of species, which is based on the polymerase chain reaction (PCR) with use of universal oligonucleotide primers (S. A. Bulat, N. V. Mironenko, 1990). PCR with isolated universal primers permits amplification of the DNA of any organism. As a result, patterns the details of which are difficult to reproduce are obtained of amplified DNA, the size and relative location of whose fragments are specific not only to a given genome, but also often to the species, strain and serotype.

#### Material and Methods

In this work we used vaccine strains of *Bac. anthracis*, STI-1; 55 (VNIIVVIM [All-Russian Institute of Veteri-

nary Virology and Microbiology]); 34F, and strains of soil saprophytes *Bac. subtilis* 433; *Bac. cereus* 8035; *Bac. pseudoanthracis* 104/2.

Chromosomal DNA was isolated by the phenol method. DNA was amplified (PCR) according to R. K. Saiki et al (1988) in 30  $\mu$ l buffer [20 mM tris-HCl; pH 7.8; 6 mM MgCl<sub>2</sub>; 10 mM (NH<sub>4</sub>)<sub>2</sub>SO<sub>4</sub>; 100  $\mu$ g/ml bovine serum albumin], containing 200 mM of each dNTP, up to 40 ng primers, 2-3 U Tag polymerase and 10-100 ng DNA.

The polymerase chain reaction was run on a thermocycler under the following conditions: DNA denaturation (93°C) for 40 s, neutralization of primers (55°C) for 40 s, DNA synthesis (71°C) for 70 s. There were 35 cycles. Amplified DNA was analyzed in 2 percent agarose gel. The universal primers were provided by the kind staff of the St. Petersburg Institute of Nuclear Physics (Gatchina).

The bacteria were analyzed using priming oligonucleotides (452; 15/45-2; 15/45-5; 21; 45).

### Results and Discussion

Any of the universal primers used initiates synthesis of DNA fragments differing in molecular weight. However, for differentiation of the bacteria we used, primers 15/45-2 and 452 were found to be the most informative.

The DNA of *Bac. anthracis* strain 55 showed a major band in the region of 770 n.p. [nucleotide pairs]. The STI-1 vaccine strain differed from strain 55 in that it had a vividly marked band in the region of 450 n.p. (Figure [not reproduced]). The soil saprophyte *Bac. pseudoanthracis* had up to seven major bands, while *Bac. cereus* had only one.

### Conclusion

The polymerase chain reaction with universal primers permits visual differentiation between *Bac. anthracis* and soil saprophytes.

### Experience in Prevention of Livestock Anthrax in Russia

957A0799B Moscow VETERINARIYA in Russian May 95 No 5, (signed to press 17 Apr 95) pp 27-30

[Article by N. G. Ipatenko, V. A. Gavrilov, A. A. Manichev, S. I. Bakhtarov, L. S. Salenko, T. N. Yakovleva, V. V. Stepanova, B. I. Shmorgun, Yu. T. Kiselev, and B. S. Sayitkulov, under the rubric "Infectious Diseases"; UDC 619:616-084:616.981.51]

[FBIS Translated Text] The mechanism of specific insusceptibility to anthrax is complicated, and it consists of a set of tissular and humoral protective factors with

marked differentiation of cellular elements. With reference to the question of mechanism of immunogenesis, use of the STI vaccine and the one derived from strain 55-VNIIVViM [All-Russian Institute of Veterinary Virology and Microbiology] established that hypodermic injection of vaccines containing 20-25 million spores per milliliter, the latter were transformed into a vegetative form in the animal. Experimental studies revealed that 3 h after injection of the vegetative form, the bacillus is demonstrable in regional and remote lymphatic nodes, as well as parenchymatous organs, where the pathogen continues to reproduce without eliciting a pathological process in the vaccinated animal. During reproduction of bacilli, there is accumulation of protective antigen that stimulates lymphoid and reticular elements of lymphatic nodes and the spleen, which causes their rapid transformation into plasma cells responsible for antibody production in vaccinated animals.

Thus, a typical immunocytic reaction develops in the animal, which is characterized by production of lymphocytes and plasma cells that synthesize protective antibodies of active phagocytes with a set of immunospecific enzymes capable of destroying encapsulated and nonencapsulated *Bac. anthracis*. There is alteration in the animal, consisting of formation of antitoxic immunity under the effect of vaccine bacilli. The accumulated neutrophils and macrophages capture vegetative and sporulated forms, digesting the former and spreading the latter in the animal. The process of assimilation of vaccine microbial cells and their transformation into complex compounds has not been entirely identified.

On the basis of experimental data we arrived at the conclusion that anthrax immunogenesis occurs in three stages. At the first stage, neutrophils (which live for several days) are involved in the reaction to administration of anthrax antigen. They interact with anthrax antigen and prepare it for subsequent effect on macrophages. At the second stage, macrophages (long-lived cells) become involved in the reaction, and they definitively prepare the vaccinated animal for exposure to T and B cells. The latter create conditions at the third stage for production of plasma cells, which are responsible for antibody formation in vaccinated animals.

At the present time 400 million head of livestock are vaccinated annually for prevention of anthrax in Russia and CIS states, using vaccine derived from strain 55-VNIIVViM. However, anthrax is recorded in different parts of Russia in the form of sporadic cases. For this reason, the question arose as to time and frequency of preventive inoculations in permanently involved regions depending on the epizootic condition of farms and regions. Analysis of statistics on regions with an anthrax problem in several oblasts, kray and

autonomous republics of Russia over the last 30-50 years revealed that in some sites the disease has been noted continuously for 10 or more years, in others — periodically at different intervals (2, 4, 6, 8 years), and in others yet — only once. Thus, in the last 50 years, anthrax was recorded in Tula Oblast once in 50.1 percent of the sites, twice in 24.5 percent, 3 times in 5.9 percent, 4 or more times in 18.5 percent of the sites. Analogous findings were made in other oblasts and krays of Russia.

The number of problematic sites with primary and secondary outbreaks of anthrax constituted 75 percent, whereas the rest of the sites where the disease was recorded 3-10 times or more, constituted 25 percent. The latter present a potential hazard to susceptible animals. On the basis of studies carried out by the Department of Veterinary Science, Russian Ministry of Agriculture and Food, proposals were submitted on time of vaccination of animals. In potentially hazardous sites (first and second category), it was recommended that cattle be inoculated in the spring or fall, depending on climate conditions and physiological condition of animals, as well as existing epizootic situation for anthrax at farms and rayons, as well as degree of danger of onset of the disease. We singled out three zones of stable, periodic and sporadic manifestation of anthrax. Preventive measures are implemented in accordance with instructions in each zone, depending on epizootic condition that has developed for anthrax in the last 10-20 years. As a result, there was dramatic improvement of the epizootic situation for anthrax in Russia.

In order to produce strong immunity in animals, it is necessary to carry out a set of organizational and explanatory measures at the farm and urban settlement prior to inoculating cattle. Kolkhoz and sovkhоз supervisors, as well as farmers, farm heads, livestock breeders, local administration together with veterinary specialists, must prepare in advance for this measure (speak on radio, lecture campaigns, announcements of date of animal inoculation) in accordance with instructions. Farm heads must assign people to build corrals, and to immobilize animals. Experience has shown that, in spite of adhering to these measures, there is mechanical omission of cattle from vaccination. For example, if the corral fences are not very high, noninoculated animals jump over them and remain unvaccinated.

The immunization document is prepared for the entire herd, and this is why sporadic cases of anthrax occur among the animals. While administering inoculations, a list is made of vaccinated animals, and it is checked against the document on livestock in the State and private sector, as well as farmers. Then, a certificate is written up in four copies for inoculated animals,

indicating the surname of the individual who gave the vaccination, date of immunization, number of inoculated animals (listed according to species), name of vaccine used and its manufacturer, series and State control numbers, date of manufacture.

After vaccination the animals are kept for about 24 h in the shade under an awning, with normal watering and feeding. During the cold time of the year, they are protected against excessive cooling. Livestock in the State and private sectors must be inoculated simultaneously. A breakdown of immunity has been observed in some vaccinated animals at a time of peak incidence of the disease (6-10 months after immunization) when appropriate upkeep and feeding conditions were not maintained.

Formation of immunity or manifestation of postvaccinal complications depended, to some extent, on the season. To this day scientists are debating about the best time to inoculate animals, spring or fall. When cattle are well-nourished in the fall, immunization is not associated with complications and produces strong immunity.

However, some animals develop temporary immunity and by the time that is the most favorable for anthrax (June-September) it is attenuated or has disappeared entirely. This is one of the causes of the sporadic cases of anthrax among vaccinated livestock. The advantage of inoculation in the spring is that the animals develop stable immunity before the start of the anthrax season. At the same time, animals with low weight at this time of the year could have postvaccinal complications, so that it is categorically forbidden to inoculate such livestock. Animals can be immunized in the spring only after they have reached an appropriate weight.

Sporadic cases of anthrax were observed in most cases in areas where there was underestimation of the importance of timely vaccination of animals at sites with permanent anthrax problems.

Immunization of animals with low resistance and low weight led to postvaccinal complications and production of brief immunity. Complications were observed when animals were kept in crowded conditions, exposed to adverse weather (heat, abrupt cooling, precipitation) and other factors (lengthy cattle drives in the broiling sun, failure to adhere to watering schedule, activation of latent infections). This was associated with dramatic decrease in resistance, the livestock presented fever, large inflammatory edema on different parts of the body, particularly at the site of inoculation, and in the cases of mixed infections (pasteurellosis, emphysematous carbuncles, enterotoxemia and bradzot of sheep, as well as parasitic diseases) there is high mortality.

In the last period some advances have been made in the study of etiology, pathogenesis, diagnosis and specific prevention of anthrax; however, the matter of its complete eradication has not yet been resolved.

In spite of the rapid accumulation of new data about anthrax, unexplainable outbreaks of the disease still occur in different parts of Russia. For example, at one of the kolkhozes, in the course of vaccinating 800 sheep, more than 100 of them jumped over a low fence and subsequently contracted anthrax. Yet the certificate indicates that the entire flock was inoculated. It is only after a thorough investigation of this case that the mistakes were discovered.

Heretofore, phage-resistant strains that circulate in nature were not taken into consideration. Of the 244 strains isolated from different zones of Russia 9 were phage-resistant. Testing for immunogenicity established that immunity to these strains was not produced with use of anthrax vaccines STI, 34F-2, 1190R and strain 55-VNIIVViM.

Practicing veterinarians are sometimes blamed for the fact that animals contract anthrax after vaccination; however, phage-resistant strains circulate in nature, and vaccines are ineffective against them.

Consequently, in the immediate future it is imperative to solve the problem of developing vaccines that would overcome phage-resistant strains.

There are also mildly virulent strains in nature, which elicit a discrete clinical form of the disease. It may not be noticed by veterinarians when they make their diagnosis, particularly in the case of abortive, latent and local forms of the anthrax process among livestock.

On 3-25 July 1992 there was an outbreak of anthrax among cattle in the Karachayevo-Cherkess Republic. As a result of forced slaughtering and processing of anthrax-ridden carcasses, 1000 people were contaminated, 21 of whom became sick and were hospitalized for the carbuncular form of anthrax (one case was fatal). Three teams of veterinary specialists were formed, and within 1 week they revaccinated the entire herd of livestock in the State and private sectors. Thanks to these steps, the outbreak of anthrax was arrested within 14 days, thus saving 280 tons of beef and mutton.

In this case, we used a new approach to arresting the infection at the site where anthrax occurred.

A specialist carried out a clinical examination and took the temperature of all livestock, with the exception of swine, at an anthrax site. A skin test with anthrax allergen was performed on swine.

The livestock were divided into two groups on the basis of clinical findings. The first group consisted of sick animals (with clinical signs of disease or elevated body temperature, as well as swine with positive reactions to anthrax allergen). This group of animals was treated with anti-anthrax serum, globulin and antibiotics. They were inoculated with anthrax vaccine 14 days after clinical recovery.

The second group consisted of animals suspected of the disease. They were vaccinated, then submitted to clinical examination and temperature measurement daily for 3 days (in the morning and evening). Animals found to have clinical signs of anthrax and a fever were switched to the first group.

Milk from the first group of animals was destroyed during the entire treatment period after decontamination by adding chlorinated lime containing at least 25 percent active chlorine (1 kg lime per 20 l milk, left for 6 h).

Milk from the second group was boiled for 3 days after vaccination and fed to animals vaccinated against anthrax, including piglets, lamb and calves. When this term elapsed, the milk was pasteurized and, under the supervision of veterinary specialists, transported through a transfer point to the assigned creamery plant for processing into butter (before quarantine was lifted). Previously, the milk was destroyed for 1 month, which resulted in a great economic loss to the farm.

No restrictions were imposed for dairy plant products from milk delivered from the farms before quarantine was imposed.

#### Conclusion

Corrections were made in the solution to the problem of using milk and implementing preventive measures at an anthrax site, as well as potentially adverse sites for this disease, with use of vaccine made with strain 55-VNIIVViM.

#### Biotechnology in Development of Methods and Ways to Diagnose Viral Infections

957A0915A Moscow VOPROSY VIRUSOLOGII  
in Russian Vol 40 No 3, May-Jun 95 pp 98-100

[Article by K. Yu. Shatalin, A. Yu. Zvonarev; UDC 616.98:578.825.16-078.33]

[FBIS Abstract] The latest discoveries in biotechnology are playing a broader role in diagnosing viral infections. This role is expected to increase as molecular biology, gene and cell engineering and immunology develop. Biotechnological methods eliminate the shortcomings of traditional approaches for obtaining viral antigens,

which include the lack of raw materials, small yield of the target product, and low quality preparations. These new methods also ensure safe working conditions and are economically profitable. The polymerase chain reaction is now receiving wide attention as a diagnostic method based on locating specific sequencing of the viral genome in the clinical samples being studied. The main advantage of the method is its high sensitivity. Recently, this method has started to play a greater role in diagnosing illnesses caused by viruses, such as the Epstein-Barr virus, the cytomegalovirus, enteroviruses, and others. Variations of this method involving the use of enzymes and other commercial systems are finding wide practical applications at the present time. 47 references.

**Stability and Viral Inhibitory Effects of Recombinant Receptorotoxins Based on Human T Lymphocyte CD4 Receptor and Diphtheria Toxin**  
**957A0915B Moscow VOPROSY VIRUSOLOGII**  
*in Russian Vol 40 No 3, May-Jun 95 pp 103-105*

[Article by A. V. Sidorov, A. G. Zdanovskiy, N. R. Shukhmina, V. V. Zverev, O. G. Andzhaparidze; UDC 616.98:578.828.6]-078.33:615.919:579.871.1]

[FBIS Abstract] Differences in the stability of CD4 receptor and diphtherial toxin-based recombinant receptorotoxins synthesized in *E. coli* are studied, as well as what causes these differences, and the differences in their primary structure. Insertion of a CD4 receptor fragment, which is responsible for HIV bonding, into the N terminus of a hybrid protein was found to lead to a drastic reduction in the stability of the hybrid polypeptide in *E. coli* and to an inability to generate a full-size protein product. *In vitro* experiments on a model of human T lymphocyte culture demonstrated that recombinant receptorotoxin stably expressed in *E. coli* inhibited the cytopathic effect of type 1 HIV and the virally induced formation of syncytium. 3 figures and 22 references.

**Role of Monoclonal Antibodies to Venezuelan Equine Encephalomyelitis Virus as Specific Activators of Immunocompetent Cells**

**957A0915C Moscow VOPROSY VIRUSOLOGII**  
*in Russian Vol 40 No 3, May-Jun 95 pp 106-109*

[Article by I. A. Razumov, T. N. Razumova; UDC 616.98:578.833.26]-07:616.153.96-097- 078.33]

[FBIS Abstract] Previous studies of the functional activity of monoclonal antibodies showed the necessity of further researching the functional activity of monoclonal antibodies and their impact on immunocompetent cells when passively introduced *in vivo*. The effects of rabbit monoclonal antiviral antibodies on the functioning

of splenic cells was studied on animals who were passively immunized. A reliable increase of the stimulation index was observed with *in vitro* activation of BALB/c murine splenocytes with the Venezuelan equine encephalomyelitis virus, the mice having been passively immunized with rat monoclonal antibodies 8D2, 3G1, and 5E7. 2 tables and 18 references.

**Mechanisms of Protective Immune Response in Monkeys with Marburg Fever**

**957A0915D Moscow VOPROSY VIRUSOLOGII**  
*in Russian Vol 40 No 3, May-Jun 95 pp 109-113*

[Article by G. M. Ignatyev, M. A. Streletsova, A. P. Agafonov, Ye. A. Kashentseva; UDC 616.98:578.824-092.9-092:612.017.1]-07]

[FBIS Abstract] Since monkeys are phylogenetically the closest to man, when Marburg fever is reproduced in monkeys, the processes which occur should most closely reflect those which occur in the human body when infected with this virus. Some indicators of immunity in monkeys both after immunization with the Marburg antigen virus and after infecting them with the homologous serum virus were studied. The parameters of nonspecific immunity (such as serum interferon production, tumor necrosis factor, and activity of natural killers), spontaneous and mitogen-induced lymphocyte proliferation, and specific immunity (antibodies to the Marburg virus and antigen-stimulated proliferation) were found to change in the course of experimental Marburg fever in immune and intact animals. The given factors were shown to influence the course of the disease. Their role in the fatal outcome of the infection was also demonstrated. 5 tables and 21 references.

**Experimental Ebola Fever in Macaca Rhesus**

**957A0915E Moscow VOPROSY VIRUSOLOGII**  
*in Russian Vol 40 No 3, May-Jun 95 pp 113-115*

[Article by O. V. Pyankov, A. N. Sergeyev, O. G. Pyankova, A. A. Chepurnov; UDC 616.98:578.833.29]-092.9-07]

[FBIS Abstract] When Macaca rhesus was aerogenously infected with the Ebola virus, a disease develops which is similar in its principal clinical and virological parameters to a grave form of the Ebola fever in humans, as it is described in the literature. The aerogenous method of infection poses particular interest, since some researchers consider this a possible route for transmittal of the infection, and it is possible that animals infected this way may not respond to treatments which are effective against other routes of infection. A rapid development of symptoms of total intoxication in the presence of a

fever, hemorrhagic diathesis, and high viremia are indicative of the severity of the infection in monkeys. 3 tables and 11 references.

**Clinical and Virological Characterization of Disease in Guinea Pigs Aerogenously Infected with Marburg Virus**

**957A0915F Moscow VOPROSY VIRUSOLOGII in Russian Vol 40 No 3, May-Jun 95 pp 119-121**

[Article by M. Yu Lub, A. N. Sergeyev, O. G. Pyankova, O. V. Pyankov, V. A. Petrushchenko, L. A. Kotlyarov; UDC 616.98:578.823.91]-092.9-036-078.59/94]

[FBIS Abstract] Guinea pigs were aerogenously infected with the Marburg virus and the dynamics of viral accumulation in the organs and tissues were studied and compared with the clinical picture of this disease. The Marburg virus (Popp strain) was found to accumulate in various organs of guinea pigs after aerogenous infection. At the initial stage of the infection, primary multiplication of the virus took place in the lungs. The presence of the virus in bronchopulmonary wipe samples two to three days after infection, and leukopenia and hyperthermia four days after infection are the earliest virological and clinical signs of disease in aerogenously infected guinea pigs. Six days after infection the virus was recorded in high concentrations in practically all organs and tissues under study. 2 tables and 7 references.

**Course of Infection in Guinea Pigs Aerogenously Infected with Venezuelan Equine Encephalomyelitis Virus**

**957A0915G Moscow VOPROSY VIRUSOLOGII in Russian Vol 40 No 3, May-Jun 95 pp 122-124**

[Article by L. Ye. Bulychev, A. N. Sergeyev, A. B. Ryzhikov, N. V. Tkacheva, Ye. I. Ryabchikova; UDC 616.98:578.833.26]-092.9-036-07]

[FBIS Abstract] The clinical, hematologic, virologic and histologic characteristics of infection in guinea pigs infected with the Venezuelan encephalomyelitis virus in aerosol form are researched. The course of viral accumulation was studied in guinea pigs aerogenously infected with the Trinidad strain of the Venezuelan equine encephalomyelitis virus. At first the agent was isolated from the lungs, and did not appear in the blood or other organs. The course of infection was characterized by viremia, multiplication of the virus, and pathomorphological changes in some of the organs and tissues of the lymphoid, hemopoietic and central nervous systems. Specifically, it affected the tonsils, lymph nodes, spleen, liver, bone marrow, some sections

of the olfactory tract and the brain. The virus was recorded in high concentrations in the brain right up until death occurred. 1 table and 9 references

**Preparation of Hyperimmune Equine Serum for Ebola Virus**

**957A0915H Moscow VOPROSY VIRUSOLOGII in Russian Vol 40 No 3, May-Jun 95 pp 138-140**

[Article by V. P. Krasnyanskiy, V. V. Mikhaylov, I. V. Borisevich, V. N. Gradoboyev, A. A. Yevseyev, V. A. Pshenichnov; UDC 615.373.3:578.833.1].012]

[FBIS Abstract] The possibility of obtaining antisera for the Ebola virus by immunizing horses and studying their properties was researched. Immunization of horses with the Ebola virus resulted in the production of specific viral neutralizing antibodies with a maximum effectiveness being attained 28 to 42 days after immunization. Repeated cycles of immunization lead to an increase of antibodies at a titer of 1:4096. Therefore, it is possible to use horses as a producer of a hyperimmune serum to the Ebola virus. A comparative analysis allows the appropriate immunization regimen which produces the maximum number of specific antibodies in the serum to be determined. 1 table and 13 references.

**Are Serotonin Receptors Receptors of Life?**

**957A0918A Moscow VESTNIK ROSSIYSKOGO AKADEMII MEDITSINSKIKH NAUK in Russian Jun 95 No 6, pp 27-30**

[Article by A. P. Simonenkov, V. D. Fedorov, A. V. Fedorov, O. S. Adrianov, Ye. A. Luzhnikov, N. P. Shugalev, M. L. Zhirnikova, V. A. Matkevich; UDC 612.018.2:577.175.823].08:612.013.1]

[FBIS Abstract] Various studies simulating primary contractions of the smooth muscles and the brain's electrical activity are described. The experiments involved using serotonin antagonists to suppress the automatic functioning and contractility of the smooth muscles and to inhibit the brain's electrical activity. Evidence is given that serotonin and serotonin receptors are a universal trigger responsible for the occurrence of primary contractions of smooth muscles, primary cardiac contractions and primary brain electric activity (EEG genesis), and for the continued maintenance of these organs and systems. It is concluded that the smooth muscles, brain and heart cannot function without serotonin and its receptors. This suggests that serotonin receptors are the receptors of life. 2 figures and 11 references.

**Yersinia Pestis Superoxide Dismutases**  
957A0918B Moscow VESTNIK ROSSIYSKOY  
AKADEMII MEDITSINSKIKH NAUK in Russian  
Jun 95 No 6, pp 45-49

[Article by O. A. Kulikov, V. I. Drobkov, I. V. Darmov, Ye. V. Smirnov; UDC 579.843.95: [579.222:577.152.191]

[FBIS Abstract] A more detailed study of the physical and chemical properties of *Y. pestis* superoxide dismutase was carried out, and some of its immunochemical and immunobiological properties were determined. Various superoxide dismutases with molecular weights of 65-67 and 200 kD have been identified in *Yersinia pestis* EV cells. These enzymes have some differences in anti-

genic specificity, and resistance to heating and inhibitors (such as cyanides and hydrogen peroxide). The activity of *Y. pestis* superoxide dismutase is a thermoinducible sign unassociated with the presence of residual plasmids of the bacilli. Superoxide dismutase can be released from the cells into a liquid culture medium. Patients inoculated with live plague vaccine develop a humoral response to high molecular-weight superoxide dismutase. Immunization of guinea pigs with purified high molecular-weight superoxide dismutase specifically ensures that they are protected after subcutaneous injection. The specific properties of *Y. pestis* superoxide dismutase make it a possible candidate for improving the diagnosis and prevention of the plague. 4 figures, 2 tables and 18 references.

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